A. M. A.
ARCHIVES OF
PATHOLOGY

OCTOBER 1953
VOLUME 56 NUMBER 4

Published Monthly by
AMERICAN MEDICAL ASSOCIATION
535 NORTH DEARBORN STREET • CHICAGO 10, ILLINOIS

Entered as Second Class Matter Jan. 20, 1926, at the Postoffice at Chicago, Under the Act of March 3, 1879. Annual Subscription, $8.00

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BIBLIOGRAPHY

1. Schilling, F. J.; De Natale, A., and Mottram, F. C.: Am. J. M. Sc. *222*:207 (Aug.) 1951.
2. Shapiro, S., and Weiner, M.: J. M. Soc. New Jersey *48*:1 (Jan.) 1951.
3. Shapiro, S., et al.: Am. Heart J. *40*:766 (Nov.) 1950.

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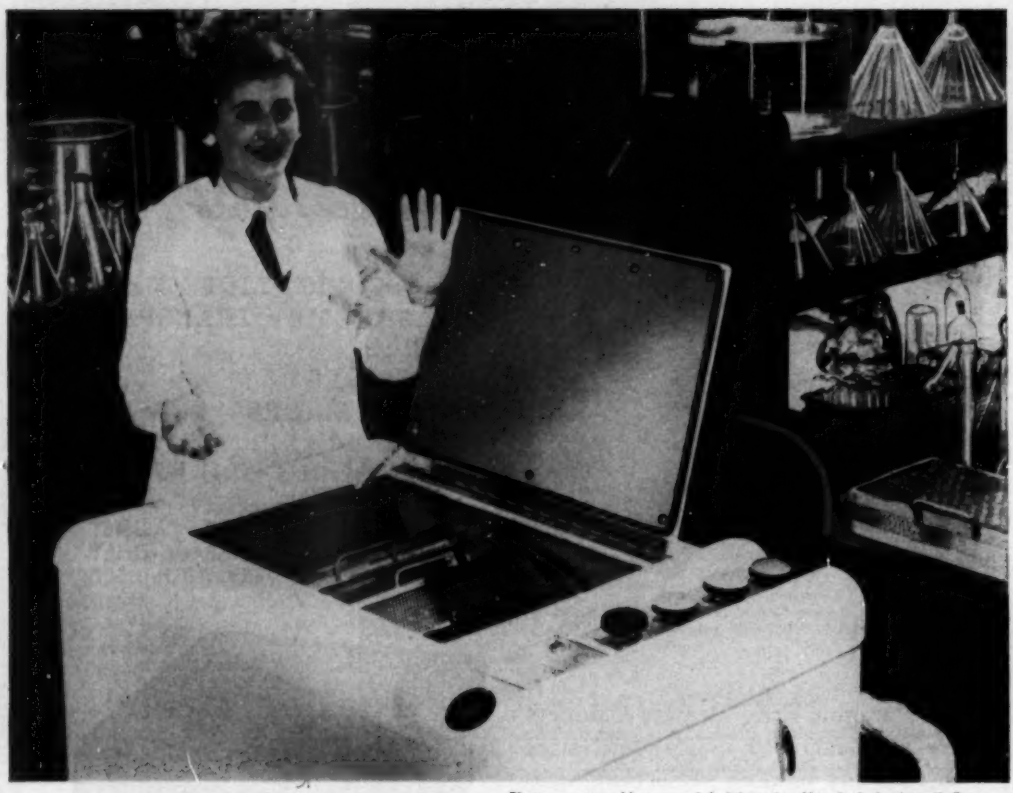

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EFFECT OF HYPERVITAMINOSIS A ON THE TESTES OF THE RAT

CHARLOTTE L. MADDOCK, Ph.D., M.D.
JONATHAN COHEN, M.D.
AND
S. BURT WOLBACH, M.D.
BOSTON

DEGENERATIVE lesions of the seminiferous tubules are among the most apparent morphological changes which accompany inanition. The proportion of tubules affected and the severity of the lesions are related to the degree of inanition and its duration. In this paper we report the occurrence of degenerative testicular lesions that are out of proportion to the nutritional states of rats receiving moderate doses of vitamin A and the inability of vitamin E to prevent these changes. During routine pathological examination of tissues [1] in an experiment on the toxic effects of long-continued administration of vitamin A, the degenerative changes in the testes were severe, while the weight gain and general condition of the animals were satisfactory. This observation plus the well-known effect of vitamin E deficiency in producing testicular lesions led to an investigation of the effect on the testicle of continued large doses of vitamin A and the influence of simultaneous administration of vitamin E.

EXPERIMENTAL

The experimental animals used were rats of Wistar strain 21 and 51 days of age. Diets employed were either Purina rat checkers or a synthetic diet previously used which gave good growth response curves. Its composition was as follows: casein (vitamin-free) 18%, dextrose 25%, cornstarch 45%, peanut oil 8%, $CaCO_3$ 1%, KH_2PO_4 2%, salt mixture [2] 1%.

The vitamin content per kilogram of diet was as follows: vitamin A 30,000 I. U. and vitamin D 5,000 I. U. (both incorporated into the peanut oil), thiamine hydrochloride 8 mg., pyridoxine hydrochloride 8 mg., riboflavin 8 mg., calcium pantothenate 16 mg., folic acid 5 mg., nicotinic acid 25 mg., paraaminobenzoic acid 300 mg., choline chloride 750 mg.

Vitamin A palmitate in sesame oil was given orally every morning, six days a weeks, at a dosage level of either 250 or 100 I. U. per gram of body weight. Vitamin E,[3] as the alpha tocopherol

This study was aided by research grants from The Nutrition Foundation and the Williams-Waterman Fund for the Combat of Dietary Disease.

From the Division of Nutritional Research, Division of Laboratories and Research, and the Department of Orthopedic Surgery, The Children's Hospital.

1. These tissues were supplied by the Hoffmann-LaRoche Company (Dr. R. Lewis), Nutley, N. J., from rats that had received 50 I. U. of vitamin A per gram of body weight for a period of one year and from dogs that had been given 25 I. U. per gram of body weight for a similar period.

2. Wolbach, S. B., and Maddock, C. L.: A. M. A. Arch. Path. **53**:273, 1952.

3. The Hoffmann-LaRoche Company supplied the vitamin A and vitamin E.

acetate, was also given orally every afternoon, six days a week, at the dosage level of 10 or 100 mg. per 50 gm. of body weight. At the lower dosage this was diluted with peanut oil. The animals were weighed daily, and their daily food intake was recorded. They were bled by cardiac puncture under ether anesthesia, killed, and organ weights were obtained immediately on an analytical balance. Postmortem examinations were performed on all animals. Tissues were fixed in Zenker's fluid and stained by Mallory's methylene blue and eosin technic. The average diameters of seminiferous tubules were obtained by determining the widths of 10 or more tubules seen in cross section on a stained fixed section of the testicle by means of a mechanical stage equipped with a vernier scale. Variations in width on any one specimen never exceeded 0.02 mm. Testicular damage was graded as follows:

±—Tubules of normal size, a rare tubule showing loss of spermatogenesis; numbers of spermatozoa decreased

+—Tubules of normal size, occasional tubules showing loss of spermatogenesis; numbers of spermatozoa decreased [4]

++—Tubules slightly decreased in diameter; frequently depression of spermatogenesis visualized; occasional tubules entirely atrophic; occasional giant cells visualized

+++—Tubules decreased in diameter; occasional areas containing mature sperm in small numbers; most tubules atrophic and containing only Sertoli cells; many giant cells visualized

++++—Spermatogenesis absent; tubules small and containing only Sertoli cells and debris

TABLE 1.—*Relation of Testicular Weight to Body Weight in Fifty-Day-Old Rats*

No. of Animal	Vitamin A, I. U./Gm. Body Weight	Vitamin E, Mg./50 Gm. Body Weight	Final Weight, Gm.	Weight of Testicles, Gm.	Testicular Injury
9 VAE	0	..	96	1.301	0
2 VAE	250	..	90	0.533	+++
6 VAE	250	..	95	1.080	++
14 VAE	250	10	94	0.692	+++
18 VAE	250	100	98	0.528	+++

RESULTS

Group 1.—One group of animals, 20 in number, was maintained on the synthetic diet. Of these 20 animals, 16 were given vitamin A at the dosage level of 250 I. U. per gram of body weight. Of these 16 animals, 8 were also given vitamin E concomitantly, 4 at 10 mg. per 50 gm. of body weight and 4 at 100 mg. per 50 gm. of body weight. The remaining four animals were used as controls for the effect of vitamin E administration only, two at the level of 10 mg. per 50 gm. of body weight and two at the level of 100 mg. per 50 gm. of body weight.

Five animals of this group were killed at 50 days. This was done to estimate progress of the experiment and to study early changes, if any, in testicular injury. Of these animals, one received vitamin E only; two, vitamin A only; one vitamin A plus vitamin E at the level of 10 mg. per 50 gm. of body weight, and one, vitamin A plus vitamin E at the level of 100 mg. per 50 gm. of body weight. These five animals are listed in Table 1 and show some interesting relationships. Body weight in this

4. Leblond, C. P., and Clermont, Y.: Ann. New York Acad. Sc. **55**:545, 1952.

group of animals varied scarcely at all, ranging from 90 to 98 gm., with the control animal occupying an intermediate position. The testicular weights are, however, much less than those of the control, those of three animals representing one-half or less of the 1.3 gm. that the testicles of the control weighed.

The remainder of the animals in this group are listed in Table 2. Control animals on vitamin E grew well, were in excellent condition when killed, and showed no

TABLE 2.—*Testicular Injury in Rats Receiving Large Doses of Vitamin A and of Vitamins A and E (Synthetic Diet)*

No. of Animal	Vitamin A, I. U./Gm. Body Weight	Vitamin E, Mg./50 Gm. Body Weight	Age at Death, Days	Final Weight, Gm.	Weight of Testicles, Gm.	Average Diameter of Tubules, Mm.	Testicular Injury
10 VAE	0	10	50	134	1.404	0.20	+
11 VAE	0	100	63	161	2.140	0.24	0
12 VAE	0	100	63	188	2.708	0.25	0
Average	...	...	..	...	2.061	0.23	.....
1 VAE	250	0	59	85	1.414	0.20	++
3 VAE	250	0	63	131	1.939	0.21	+
4 VAE	250	0	62	121	1.916	0.23	+
5 VAE	250	0	63	104	1.880	0.21	+
7 VAE	250	0	56*	95	1.649	0.19	+
8 VAE	250	0	58*	137	1.983	0.24	±
Average	...	...	..	...	1.798	0.21	.....
13 VAE	250	10	63	108	0.692	0.14	+++
15 VAE	250	10	62*	92	0.796	0.16	+++
16 VAE	250	10	58*	147	2.213	0.24	0
17 VAE	250	100	59*	91	0.512	0.14	+++
19 VAE	250	100	62*	116	0.425	0.14	+++
20 VAE	250	100	62*	125	0.700	0.14	+++
Average	...	...	..	...	0.889	0.16	.....

* Died.

TABLE 3.—*Testicular Injury in Rats Receiving Large Doses of Vitamin A (Stock Diet)*

No. of Animal	Vitamin A, I. U./Gm. Body Weight	Vitamin E, Mg./50 Gm. Body Weight	Age at Death, Days	Final Weight, Gm.	Weight of Testicles, Gm.	Average Diameter of Tubules, Mm.	Testicular Injury
1 AK	250	0	88*	73	0.787	0.13	++++
2 AK	250	0	99†	110	1.446	0.17	++++
3 AK	250	0	114†	185	1.320	0.22	++++
4 AK	250	0	114†	205	1.305	0.18	++++
5 AK	250	0	114†	208	2.287	0.23	++++
6 AK	250	0	99†	128	1.174	0.20	++++

* Died.
† Killed.

pathologic changes in any of the tissues examined. Among those animals receiving only vitamin A, two died of hemorrhage; one was in poor nutritional state, and one showed transient signs of a hemorrhagic tendency, which was absent at the end of the experiment. This one, as well as the two remaining rats, was in reasonably good condition and would undoubtedly have lived for a longer period. Testicular injury ranged from ± in one, 1 + in four, to 2 + in one. The two 50-day animals in this category showed testicular changes of 2 + and 3 +.

In the combined vitamin A and E subgroup, there were five moribund animals showing multiple and massive hemorrhages. The sixth animal also showed hemorrhage in one hindleg but would probably have survived a few more days. One animal which died because of hemorrhage had normal testicles; the rest showed maximum or 3 + injury. The two 50-day animals in this group also showed 3 + injury. There appeared to be no relationship between the hemorrhagic syndrome and the testicular change.

Group 2.—Six weanling rats maintained on Purina rat checkers were started on doses of vitamin A at the level of 250 I. U. per gram of body weight. The results are given in Table 3. The first animal in this series died apparently of toxic effects of vitamin A administration and not of hemorrhage. Healed fractures of the tibiae

TABLE 4.—*Testicular Injury in Rats Receiving Large Doses of Vitamin A (Synthetic Diet)*

No. of Animal	Vitamin A, I. U./Gm. Body Weight	Vitamin E, Mg./50 Gm. Body Weight	Age at Death, Days	Final Weight, Gm.	Weight of Testicles, Gm.	Average Diameter of Tubules, Mm.	Testicular Injury
38 VAE.....	100	0	135*	173	2.464	0.29	++
39 VAE.....	100	10	100*	170	2.250	0.22	++
41 VAE.....	100	100	75*	140	2.525	0.24	++
42 VAE.....	100	100	75	176	2.353	0.23	++

* Died.

TABLE 5.—*Testicular Injury in Rats Receiving Large Doses of Vitamin A (Stock Diet)*

No. of Animal	Vitamin A, I. U./Gm. Body Weight	Vitamin E, Mg./50 Gm. Body Weight	Age at Death,* Days	Final Weight, Gm.	Weight of Testicles, Gm.	Average Diameter of Tubules, Mm.	Testicular Injury
49 VAE.....	100	0	95	272	2.930	0.27	0
51 VAE.....	100	10	103	351	3.169	0.23	+
53 VAE.....	100	10	95	338	3.423	0.28	+
54 VAE.....	100	10	93	372	3.567	0.24	+
55 VAE.....	100	100	95	325	2.920	0.28	+
56 VAE.....	100	100	93	267	2.918	0.23	+
57 VAE.....	100	100	93	347	2.994	0.27	+
58 VAE.....	100	100	93	332	3.004	0.27	+
59 VAE.....	100	100	60	256	2.284	0.26	+
60 VAE.....	100	100	103	311	2.846	0.25	+

* Killed.

were present. The other animals were in a state of poor to moderate nutrition, showing the bony changes (recent and healed fractures, the consequence of accelerated remodelling processes at the growing ends of bone [5]) characteristic of excessive vitamin A intake. The testicles were small to moderate in size, and all showed advanced degenerative changes.

Group 3.—Because of failure of most of the animals in the previous group to survive beyond 63 days (representing approximately 26 doses of vitamin A) on combined vitamins A and E at the vitamin A level of 250 I. U. per gram of body weight, vitamin A intake of this group was lowered to 100 I. U. per gram of body weight. Four animals on the synthetic diet were maintained on this regimen, and

5. Wolbach, S. B.: J. Bone & Joint Surg. **29**:171, 1947.

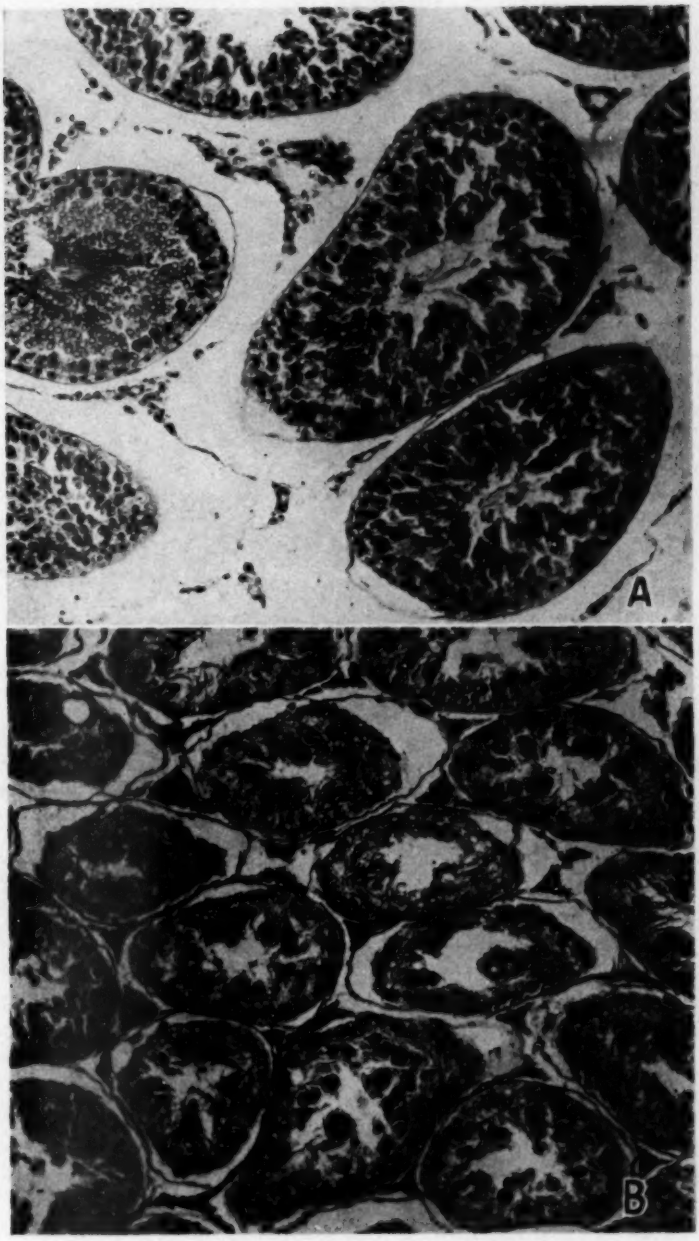

Fig. 1.—*A*, testis of 100-day-old rat raised on a synthetic diet and given vitamin A in 51 doses of 100 I. U. per gram of body weight and vitamin E at 10 mg. per 50 gm. of body weight starting at 21 days of age. There is clumping of the spermatids and spermatocytes and loss of chromatin of the spermatocytes. There is moderate edema between tubules; × 125. *B*, testes of 88-day-old rat raised on stock diet and given 44 doses of vitamin A at 250 I. U. per gram of body weight starting at the age of 27 days. The tubules are much smaller than in Figure 1*A*. They all show profound degenerative changes; × 125.

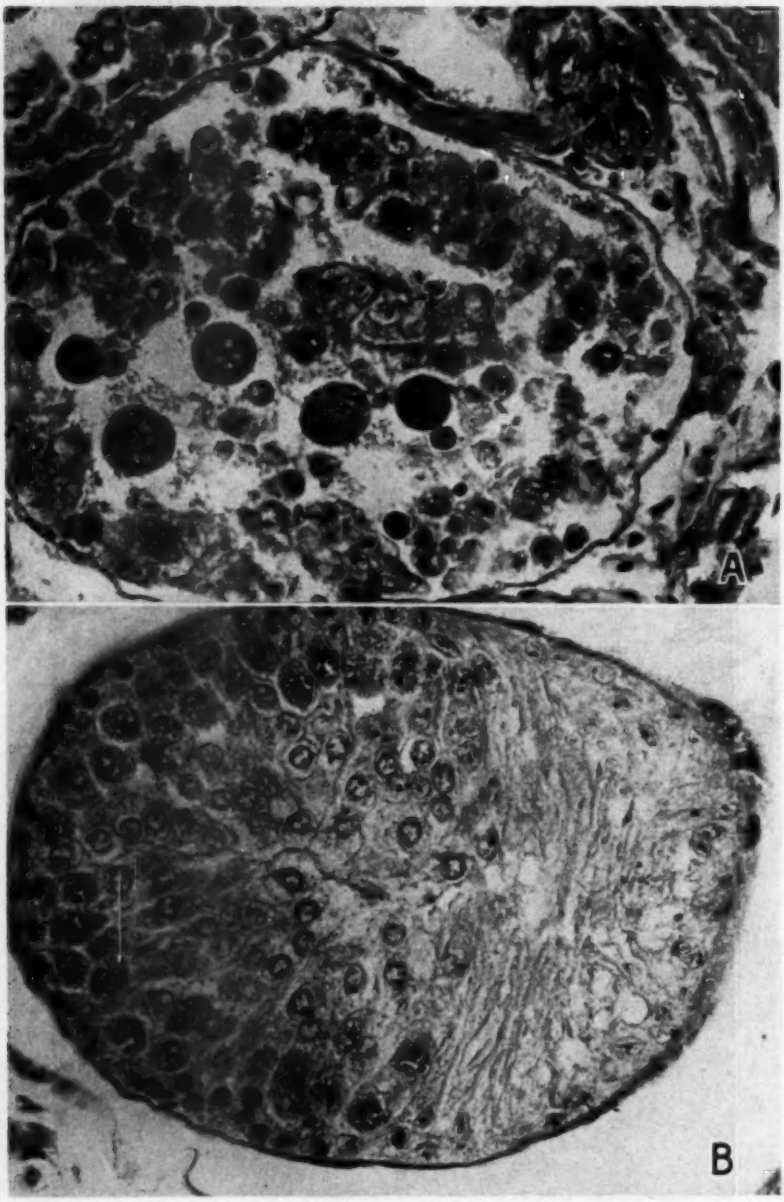

Fig. 2.—*A*, one tubule of same animal as in Figure 1*B*, showing details of giant cell formation and degeneration of spermatocytes; × 400. *B*, one tubule from a rat aged 99 days, raised on a stock diet, and given 54 doses of vitamin A at 250 I. U. per gram of body weight starting at age of 27 days. This shows the progressive degeneration of spermatocytes and spermatogonia; × 400.

survival was prolonged from 75 days to 135 days (the upper limit occurring in the non-vitamin-E-supplemented animal), but death from hemorrhage again supervened. Testicular change was designated as 2 + in all four animals. The results are given in Table 4.

Group 4.—Ten weanling rats on the stock diet were maintained on the vitamin A level of 100 I. U. per gram of body weight, three being given vitamin E at 10 mg. per 50 gm. of body weight and six at the 100 mg. per 50 gm. of body weight. All were killed at periods varying between 80 and 103 days, and all were in an excellent state of nutrition. The testicles showed no gross changes or diminution in size, but on histological examination all of them, with the exception of the one rat on vitamin A only, showed minimal testicular change. Results are given in Table 5.

Group 5.—Observations were made on the effect of long-maintained large doses of vitamin A on the mature rat to note the action of the vitamin upon the fully developed testicle. Five 51-day-old sexually mature rats that had been maintained on a stock diet since weaning were given vitamin A at two levels, three at 250 I. U. per gram of body weight and two at 100 I. U. per gram of body weight. One rat on each vitamin A dosage also received daily supplements of vitamin E, 100 mg. per 50 gm. of body weight. After a 100-day period on this regimen the rats were killed. No evidence of testicular injury could be demonstrated either in the straight vitamin A category or in that supplemented with vitamin E.

The histologic characteristics of the testicular degeneration in the early stage consisted of a diminution in the numbers of spermatozoa and a loss of chromatin by the spermatids and spermatocytes (Fig. 1*A*). Late lesions contained spermatocytes with bead-like nuclei with diminished chromatin, as described by Mason,[6] and in the more advanced lesions there were sloughing of the spermatocytes and clumping of their nuclei and the nuclei of spermatids to form giant cells (Figs. 1*B* and 2). At the end stage nearly complete sloughing of the germinal epithelium occurred. While the changes seen are thought to be nonspecific and to conform in general to those elicited in inanition, the chromolytic nuclear changes and the giant cell formation resemble the changes described in vitamin E deficiency. The reversibility of the lesion was not investigated.

COMMENT

It seems fairly clear from the data submitted and from evidence on hand from the long-continued toxicity experiments on vitamin A conducted in the Hoffmann-LaRoche Laboratories, as well as rather fragmentary observations made on previous animals subjected to the hypervitaminosis A regimen, that testicular injury is one of the results of the prolonged administration of large doses (50 to 250 I. U. per gram of body weight) of this vitamin. It would also appear that this effect is potentiated by the concurrent intake of vitamin E. An explanation of this enhancing effect of vitamin E may reside in its antioxidant action, which presumably occurred in the blood stream or tissues since the two vitamins were administered at different times. Our data are not sufficiently extensive or inclusive to assess what part, if any, diet plays in this picture, but there is an indication that on the synthetic diet the rats were more prone to development of the testicular lesions.

6. Mason, K. E.: Am. J. Anat. **52**:153, 1933.

The complicating factor of hemorrhage, which apparently occurs earlier and with greater regularity in our rats on synthetic diet, makes it impossible to carry on the experiment for equally long periods of time at equivalent levels of vitamin A in the two diets used, synthetic and stock. One of the most interesting features of the study, one that will be reported in a subsequent paper, has been the intensification of the hemorrhagic diathesis induced in hypervitaminosis A by the simultaneous administration of vitamin E.

SUMMARY

Long-continued ingestion of moderate doses of vitamin A in weanling rats elicits degenerative lesions of the testis, but in mature rats receiving similar dosages these lesions do not develop.

Vitamin E given concurrently with vitamin A potentiates the degenerative change.

GERMINAL NATURE OF TERATOID TUMORS OF THE THYMUS

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THE OCCURRENCE of teratoid tumors in the mediastinum is well known. Autopsies were held on two patients who had suffered from such growths, and the tissues were studied in the light of recent work. The findings indicated that certain mediastinal teratoid tumors are unequivocally extragenital, probably thymic and possibly germinal.

In one of the two cases studied, a boy of 16 died of metastases from a teratocarcinoma of the mediastinum. By multiple block serial sectioning of the testes a gonadal primary tumor was excluded, and the mediastinal growth was found to originate in, or adjacent to, the thymus. Although the presence of trophoblastic tissue characterized both the primary tumor and the metastases, a nodule of germinomatous tissue was found in immediate proximity to thymic tissue. The second patient was a man of 37 who died with generalized chorioepitheliomatosis apparently originating in the mediastinum. The testes were eliminated as a possible primary site by the multiple block technique. The presumably primary mediastinal growth arose in, or adjacent to, the thymus.

REPORT OF CASES

CASE 1.—A 16-year-old white boy was admitted to the hospital with the complaint of chest and abdominal pain of three weeks' duration. The pain began as mild epigastric distress; soon the entire abdomen was painful, and dyspnea and fever were noted. At first the complaints were not disabling, and he continued at school for five days, after which he remained at home. One week before admission to the hospital he complained that a dull pain in the left side of his chest was aggravated by respiration. No cough or hemoptysis were noted. He lost about 10 lb. (4.5 kg.) in weight in the two weeks before admission.

The past history revealed no illness other than frequent upper respiratory infections and bronchial asthma. A chest survey film taken six months before had been reported as negative, but review of the film revealed a slight enlargement in the left pulmonic area.

Physical Examination.—The boy appeared acutely ill and showed signs of recent weight loss. Small cervical nodes were palpable on the left side of the chest. There was dullness to percussion over the entire left side of the chest, and the breath sounds were decreased. The diaphragm on the left was fixed, and the respiratory excursion of the rib cage on this side was decreased. The right heart border was displaced 2 cm. to the right. The cardiac rhythm was regular, and the sounds were slightly muffled. The blood pressure was 104/58, and the pulse rate was 104 per minute; respiration was increased to 22 per minute. There was considerable abdominal rigidity, but no masses or organs were palpated. Axillary lymph nodes were small and shotty.

From the Division of Laboratories, Cedars of Lebanon Hospital.

Laboratory Data.—The red blood cell count was 4,300,000, and the hemoglobin was 12 gm. per 100 cc. The white blood cell count was 10,000, with a differential count of 74% polymorphonuclear leucocytes, 8% eosinophiles, 17% lymphocytes, and 1% monocytes. The sedimentation rate (corrected) was 28 mm. in one hour (Wintrobe). The urine was negative for albumin and sugar. The specific gravity was 1.025. The sediment was negative. Blood sugar was 98 mg. per 100 cc. Sputum smear, culture, and guinea-pig inoculations were negative for acid-fast bacilli.

X-ray examination of the chest revealed a considerable amount of fluid in the left pleural cavity, with a fairly marked shift of the heart and mediastinum to the right.

Course.—After admission to the hospital approximately 1,200 cc. of serosanguinous fluid was removed from the left chest. It showed red blood cells and leucocytes, but no bacteria were seen on smear or culture. The Ziehl-Neelsen stain and cultures for tubercle bacilli were negative.

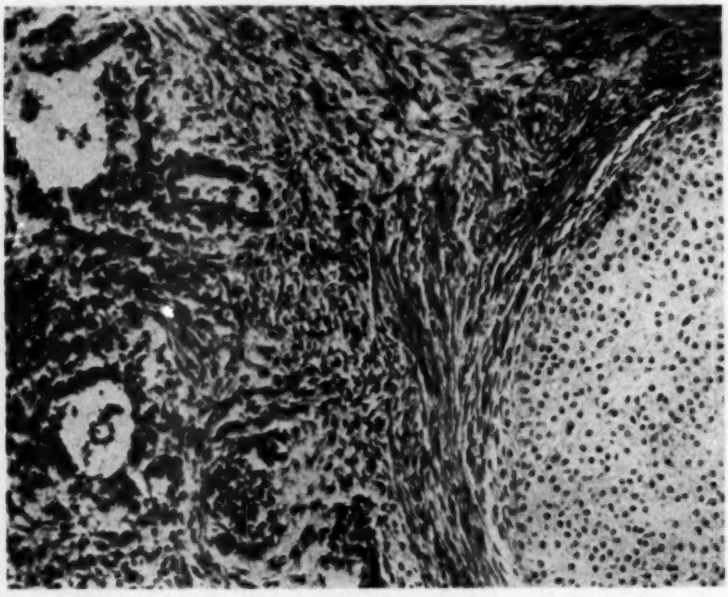

Fig. 1 (Case 1).—Teratocarcinoma of the mediastinum; × 80.

Smears disclosed no malignant cells. Some improvement in respiration was noted following the thoracentesis, but the abdominal pain persisted. A tuberculin skin test was negative. Bronchoscopy revealed compression and torsion of the left main bronchus. Spontaneous pneumothorax, tuberculosis, and neoplasm were considered in the differential diagnosis. The patient continued to be febrile, and the afternoon temperature rose to between 102 and 103 F. daily.

An exploratory thoracotomy was done, and the entire pleural cavity on the left was found to be filled with tumor and clotted blood. The mass extended from the midaxillary line to the anterior chest wall and from the ribs to the pericardium. It measured about 30 cm. in diameter and was extremely vascular. The left lung was compressed and covered by a thick membrane. A 13 cm. mass of tumor could be removed by blunt dissection, and the membrane was removed from the pleura, allowing partial reexpansion of the lung. A pneumonectomy was not considered feasible.

Examination of the surgical specimen revealed a 13 cm. ragged and focally hemorrhagic variegated lobulated mass with cystic areas containing mucoid material and yellow to glistening

gray areas surrounded by foci of hemorrhage. Microscopic sections showed (Fig. 1) a loose vascular matrix containing many glandular structures lined by cuboidal or columnar epithelium showing prominent cytoplasmic vacuolization. Other areas showed immature muscle, cartilage, and spaces lined by respiratory epithelium. Areas of nerve and glial tissue could be recognized. Multinucleated cells resembling syncytial trophoblast were seen. A diagnosis of teratocarcinoma of the mediastinum was made.

Postoperatively the temperature declined to an afternoon maximum of 99 to 100 F. X-rays showed a large mass of tumor remaining, with the heart still displaced to the right. The urine was tested for gonadotropins (Rana pipiens frog test), with negative results. Progress was satisfactory, and the patient was released from the hospital two weeks after the operation.

The patient was readmitted to the hospital one month later because of pain, dyspnea, weakness, and additional weight loss. He showed increased pallor, and the red blood cell count was found

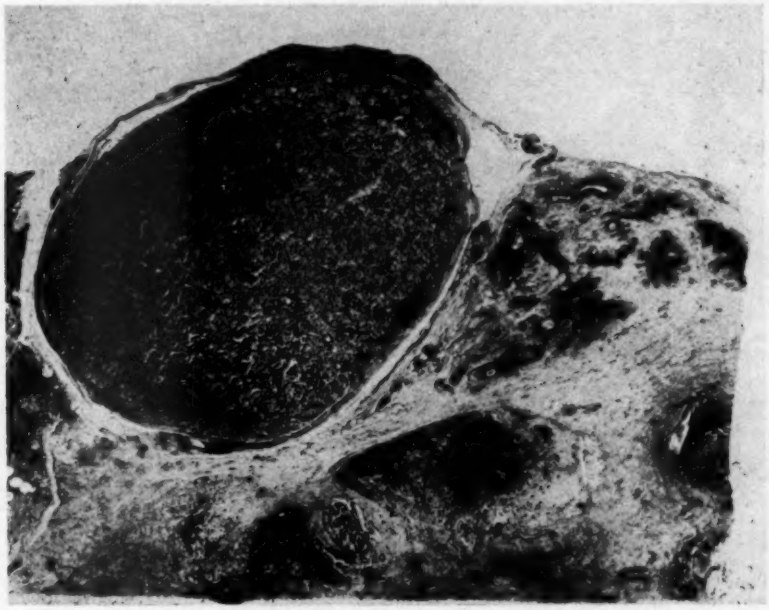

Fig. 2 (Case 1).—Teratocarcinoma of the thymus. Note the hemorrhagic teratocarcinomatous portion, the residual thymic tissue, and the germinomatous nodule; × 6.

to have dropped to 2,500,000, with 5.4 gm. of hemoglobin per 100 cc. The liver was palpable 4 cm. below the costal margin, and the spleen was questionably enlarged. Five pints (2.4 liters) of blood was given within a two-day period. Ten millicuries of radioactive (P^{32}) colloidal chromic phosphate was injected into the left pleural cavity to control the formation of fluid. The patient was discharged but died at home four days later.

Autopsy Findings.—The left pleural cavity was almost completely filled with a firm red-tan tumor mass which was adherent to the chest wall except at the site of the previous surgery. The mass filled the anterior mediastinum and extended to the midaxillary line. It centered in the region of the thymus, displaced the heart to the right, and compressed the left lung. The tumor measured 30 cm. in diameter and with the attached left lung weighed 2,540 gm. Microscopic examination yielded findings resembling those seen in the surgical specimen, and many areas of embryonic tissue with glands and cartilage were seen amid areas of hemorrhage and necrotic tumor. Compressed thymic tissue with prominent Hassall's corpuscles was found overlying the

tumor, and in an area (Fig. 2) adjacent to both thymic and tumor tissue was a nodule 1 cm. in diameter which exhibited a classic germinomatous pattern (Fig. 3).

The heart and great vessels were negative. The lungs were not abnormal except for atelectasis. The liver was enlarged and weighed 2,500 gm.; microscopically, metastatic teratocarcinoma was seen. The spleen weighed 220 gm. and contained several tumor nodules within the pulp; sections showed extramedullary hemopoiesis as well. The bone marrow revealed tumor invasion and hyperplasia of the remaining marrow. The thyroid, adrenals, kidneys, pituitary, and brain were not involved.

The testes were fixed, and the entire parenchyma was cut into blocks 0.4 cm. thick. Sections were cut from each block. Neither neoplastic tissue nor scars were present.

Case 2.—A 37-year-old white man was admitted to the hospital with complaints of cough, anorexia, and pain in the right scapula. He had been apparently well until six months before,

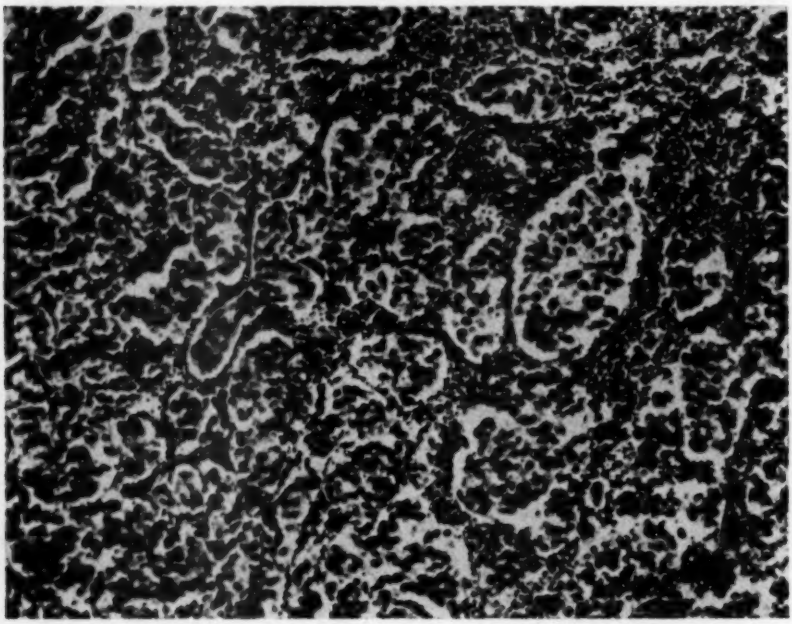

Fig. 3 (Case 1).—Germinomatous tissue in teratocarcinoma of the thymus; × 120.

at which time a nonproductive cough was noted, with anorexia. Pain in the right scapular region began two months before admission and radiated to the neck and lateral aspect of the right arm. In the six months before admission he lost 30 lb. (13.6 kg.) and complained of increasing weakness and fatigability. He had no fever, night sweats, or hemoptysis. Six weeks prior to admission he began to have substernal discomfort. A diagnosis of mediastinal tumor was made elsewhere, and a course of deep x-ray therapy had been given three weeks before admission. Enlargement of the tumor mass was noted during the period of therapy.

Physical examination was essentially negative. The blood pressure was 140/80, and temperature, pulse, and respiration were normal. The genitalia showed no testicular enlargement or tenderness.

Laboratory Data.—The red blood cell count was 4,700,000, and the blood contained 12.5 gm. of hemoglobin per 100 cc. The white blood cell count and the differential count were normal. The urine had a specific gravity of 1.025, and the sediment was negative. It contained no sugar or albumin.

X-ray examination of the chest revealed a lobulated tumor mass in the right side of the anterior mediastinum measuring about 15 cm. in length and 7 cm. at its widest point.

Course.—Exploratory thoracotomy was done on the third day of hospitalization. The mediastinum, except for the superior portion, was found to be occupied anteriorly by a large mass, 15 cm. in diameter, which was contiguous with the body of the thymus gland. In the lower portion the mass invaded the middle lobe of the right lung, and at this point it was 2 in. (5 cm.) in diameter. A chain of walnut-sized lymph nodes was evident along the trachea. The large mass infiltrated between the hilar vessels and surrounded the superior vena cava, compressing the innominate vein. The thymus was adherent to the inferior thyroid vein, which was markedly distended. There was a very large subcorynal node and a large node just above the diaphragm slightly compressing the inferior vena cava. The vessels over the pericardium were markedly enlarged and hyperemic. With blunt and sharp dissection a portion of the tumor was removed. A nodule of tumor tissue was palpated in the lower lobe of the right lung, and a similar mass, approximately 2 cm. in diameter, was felt in the upper lobe. The diaphragmatic surface on the right was irregular and thought to indicate tumor involvement of the underlying dome of the liver. A markedly enlarged thoracic duct was seen distended with chyle. The patient received 1,500 cc. of whole blood during the operation and was in good postoperative condition.

Examination of the removed tumor shows an 11 cm. soft lobular mass with a mottled tan and red surface. There were foci of hemorrhage, and the tumor grossly resembled placental tissue. Microscopic examination showed a cellular tumor made up of huge pleomorphic cells having an abundant clear to slightly eosinophilic granular cytoplasm. The nuclei demonstrated extreme hyperchromatism and pleomorphism. Many cells were multinucleated and resembled syncytiotrophoblasts. The tumor was extremely vascular and had large areas of hemorrhagic necrosis. Around the edges, compressed thymic tissue was found. Many lymph nodes showed complete replacement by neoplastic tissue. A diagnosis of chorioepithelioma of the thymus was made.

On the second postoperative day 2μc of P^{32} was administered intrapleurally on the right. Jaundice was noted clinically on the same day, and the icteric index was 29.3 units. On the fourth postoperative day 3μc of P^{32} was again injected into the right thoracic cavity. The testes were reexamined and found negative. Gonadotropic activity was found in the urine (Rana pipiens frog test). One week after surgery the red blood cell count was 3,880,000, with 9.7 gm. hemoglobin per 100 cc.; the white blood cell count, 9,700, with 78% polymorphonuclear leucocytes, 0.5% basophiles, 7.5% lymphocytes, 14% monocytes. Urinary urobilinogen rose to the dilution of 1 : 40. Blood cholesterol was 192 mg. per 100 cc.; cholesterol esters were 74 mg. per 100 cc.; the icterus index was 21.4 units and thymol turbidity 2 units. The cephalin flocculation test was negative. A new metastatic nodule in the left lung was seen on a postoperative x-ray. On the 13th postoperative day the general condition of the patient had deteriorated, and therapy with cortisone acetate was begun in daily 100 mg. intramuscular doses. Urine showed 1+ albumin, was positive for bile, and contained a few white blood cells and casts. Red blood cells were 2,970,000, with 7.3 gm. hemoglobin per 100 cc.; white blood cells were 22,200, with 85% polymorphonuclear leucocytes, 7.5% lymphocytes, 5.5% monocytes, 0.5% myelocytes, 1.5% metamyelocytes. Three nucleated red blood cells were seen in a count of 200 white blood cells. The icterus index rose to 41.5 units. The patient failed to respond to cortisone, continued downhill, and died on the 17th postoperative day. The temperature during the whole period of hospitalization showed an early morning rise to about 101 F. The pulse rate varied between 100 and 120 per minute.

Autopsy Findings.—The right pleural space was filled with bloody fluid, and in the anterior mediastinum there was a dark hemorrhagic tumor mass adherent to the sternum and infiltrating the right third rib. The tumor appeared to arise from the region of the thymus, and compressed thymic tissue was seen surrounding it. Microscopic examination of the neoplasm showed the same pattern of pleomorphic cells resembling trophoblast as was seen in the surgically removed tumor. The lungs were infiltrated by tumor nodules varying from 0.5 to 2.0 cm. in diameter. The paratracheal, peribronchial, and mediastinal nodes were involved by neoplastic tissue. The

3,800 gm. liver and 840 gm. spleen were riddled by tumor nodules from 3 to 5 cm. in diameter. There also were metastases to the pancreas, adrenals, kidneys, thyroid, brain, bones, heart, aorta, stomach, and intestines.

The testes were grossly normal, weighed 20 gm. each, and were completely serially blocked, as in Case 1. The microscopic sections revealed no tumor or scars; only atrophy of the tubules and Leydig cell hyperplasia were seen.

COMMENT

Schlumberger [1] pointed out the relationship of mediastinal teratomas to the thymus and devised an ingenious theory which accounted for the formation of these growths by derivation from the thymic anlage. Supporting his observation in the two present cases, thymic tissue was found intimately related to the presumably primary growth. The importance of the study of the thymus in cases of intrathoracic teratoid growth is not always appreciated, as in the case recently reported by Kay and Reed.[2] A number of workers have cautioned against the acceptance of extragenital teratoid tumors in men unless neither neoplastic tissue nor a cicatrix indicating regression of a tumor could be located in the testes. In the two present cases a testicular primary growth was excluded by multiple block sections of the entire testicular parenchyma.

The observation of Willis [3] that the more malignant trophoblastic teratoid tumors of the mediastinum occurred more commonly in men than in women was borne out by the nature of the growths in our patients. The question of the cell of origin and the morphogenetic sequences in teratoid tumors has not been entirely settled, even for testicular tumors, and in the case of extragenital neoplasms, such as those of the thymus, there is even more difference of opinion. The theory of evolvement from a neoplastic equivalent of the primordial germ cell suggested by Friedman [4] is not supported by the failure to find germinomas among testicular tumors in children [5] despite the presence of the other well-accepted types of growth. Dixon and Moore [6] have suggested, as have others, that the germinoma represents a sideline of development and is not in the direct line of differentiation from the cell of origin to the more complex teratoid tumors. If one does not accept the germinoma of the testis as originating from primordial germ cells, the seminiferous epithelium provides the only alternative tissue of origin. The occurrence of germinomatous tissue in the thymus (and pineal gland), however, could hardly be accounted for on such a basis. We can offer no satisfactory explanation for the localization of primordial germ cells in the thymic anlage. Further evidence is available from embryologic studies.

1. Schlumberger, H. G.: Teratoma of the Anterior Mediastinum in the Group of Military Age: A Study of 16 Cases, and a Review of Theories of Genesis, Arch. Path. **41**:398-444, 1946.

2. Kay, S., and Reed, W. G.: Chorioepithelioma of the Lung in a Female Infant 7 Months Old, Am. J. Path. **29**:555-567, 1953.

3. Willis, R. A.: Atlas of Tumor Pathology: Section 3, Fascicle 9, Teratomas, Washington, D. C., Armed Forces Institute of Pathology, 1951.

4. Friedman, N. B.: The Comparative Morphogenesis of Extragenital and Gonadal Teratoid Tumors, Cancer **4**:265-276, 1951.

5. Mostofi, F. K.: Testicular Tumors in Infants, Abstracted, Am. J. Path. **28**:536, 1952.

6. Dixon, F. J., and Moore, R. A.: Atlas of Tumor Pathology: Section 8, Fascicles no. 1 31b and 32, Tumors of the Male Sex Organs, Washington, D. C., Armed Forces Institute of Pathology, 1951.

Although Witschi [7] expressed doubts as to the relationship of embryomata to germ cells, except for some gonadal ones, the cells illustrated in his study of the migration of primordial germ cells in human embryos certainly resemble the cells of germinomas.

SUMMARY

In two patients with teratoid tumors of the mediastinum the primary tumor was localized in the region of the thymus. The testes were excluded as the primary site by means of multiple block serial sections of the testes. In one of the two primary tumors a nodule of germinomatous tissue was found. These observations support the view that mediastinal teratoid tumors arise in the thymus and that the teratomas and chorioepitheliomas of the thymus may evolve, as in the comparable tumors of the testis, from a tumor of primordial germ cells.

7. Witschi, E.: Migration of the Germ Cells of Human Embryo from the Yolk Sac to the Primitive Gonadal Folds, Contrib. Embryol. **32**:69-80, 1948.

NECROTIZING NEPHROSIS IN THE RAT FOLLOWING ADMINISTRATION OF CARBON TETRACHLORIDE

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THE RECENT literature contains many case reports of fatal carbon tetrachloride (CCl_4) poisoning in humans.[1] The renal lesion usually described in these cases is a so-called lower nephron nephrosis. No comparable lesion, however, has been reported to follow carbon tetrachloride intoxication in experimental animals, and, in addition, a careful search of the literature reveals no well-documented studies of the renal injury produced by single doses of this chlorinated hydrocarbon. Therefore, the following study was made of the effect on the kidney of single doses of carbon tetrachloride administered subcutaneously and intraperitoneally to rats. It was found that carbon tetrachloride administered intraperitoneally was highly lethal, producing a striking bilateral necrosis of the proximal tubular cells. In contrast, carbon tetrachloride administered subcutaneously in comparable dosage was not lethal and caused no detectable renal lesion.

EXPERIMENTS

Male and female albino rats of mixed laboratory stock were used. They were fed rat chow,[2] allowed tap water ad libitum, and kept in individual cages in air-conditioned rooms at 70 F. Ninety-four rats were used, divided into three groups.

GROUP 1.—Group 1 consisted of 21 animals which were given chemically pure carbon tetrachloride (Merck & Company) intraperitoneally (Table 1).

From the Department of Pathology, Northwestern University Medical School, and Wesley Memorial Hospital.

1. (*a*) Lucke, B.: Lower Nephron Nephrosis (the Renal Lesions of the Crush Syndrome, of Burns, Transfusions, and Other Conditions Affecting the Lower Segments of the Nephrons), Mil. Surgeon **99**:371-396, 1946. (*b*) Dunn, J. S.; Gillespie, M., and Niven, J. S. F.: Renal Lesions in 2 Cases of Crush Syndrome, Lancet **2**:549-552, 1941. (*c*) Mallory, T. B., editor: Case Records of the Massachusetts General Hospital, Case 34221, New England J. Med. **238**:776, 1948. (*d*) Woods, W. W.: The Changes in the Kidneys in Carbon Tetrachloride Poisoning, and Their Resemblance to Those in the "Crush Syndrome," J. Path. & Bact. **58**:767-773, 1946. (*e*) Smetana, H.: Nephrosis Due to Carbon Tetrachloride, Arch. Int. Med. **63**:760-777, 1939. (*f*) Moon, H. D.: The Pathology of Fatal Carbon Tetrachloride Poisoning, with Special Reference to the Histogenesis of the Hepatic and Renal Lesions, Am. J. Path. **26**:1041-1057, 1950. (*g*) McManus, J. F. A., and Rutledge, G. L., Jr.: "Lower Nephron Nephrosis" a Misnomer for the "Crush" Kidney, ibid. **25**:771, 1949.

2. Rockland rat diet (pellet form) consists of 21% protein, 4% fat, 6% fiber, the remainder consisting of carbohydrate plus minerals and vitamins. The vitamins include thiamine, 228 to 465 γ per 100 gm.; riboflavin, 583 to 652 γ per 100 gm.; pyridoxine, 209 to 275 γ per 100 gm.; pantothenic acid, 1,989 to 2,568 γ per 100 gm.; vitamin A, 610 to 680 I. U.; vitamin D, 148 U. S. P. units; alpha tocopherol, 7 mg. per 100 gm.

GROUP 2.—Group 2 consisted of 45 rats which were given varying doses of chemically pure carbon tetrachloride subcutaneously in one or both flanks (Table 2).

GROUP 3.—Group 3 consisted of 26 rats which were used as controls and received no carbon tetrachloride. Included in this group were 12 animals on a high fat-low protein diet consisting of vitamin-free casein 8%, Crisco 35%, sucrose 53%, salt mixture No. 2 4%. A daily vitamin supplement consisted of thiamine 25 γ, riboflavin 40 γ, pyridoxine 25 γ, calcium pantothenate 100 γ. A weekly supplement consisted of vitamin A 6,300 I. U., vitamin B 900 I. U., alpha tocopherol 1 mg. (10 gm. of this mixture was allowed each rat per day).

Autopsies were performed promptly after the animals died or had been killed by a blow to the head. Pieces of kidney, liver, lung, heart, adrenal gland, spleen, stomach, and gonad were routinely fixed for 24 hours in 10% buffered formalin and in Helly's solution. With some animals, sections of liver and kidney were studied after freezing dehydration.[3] The blocks were embedded in paraffin, and sections were cut at 5 μ and stained with hematoxylin and eosin.

TABLE 1.—*Data on Carbon Tetrachloride Administered Intraperitoneally*

Animal No.	Sex	Diet*	Weight, Gm.	CCl_4/100 Gm. Body Weight, Cc.	Hr. Alive†	P.T. Nec.‡	Zone of Necrosis	Hb.-Pos. Droplets in P.T.§	Liver C.L. Nec.‖
14	M	N	218	0.20	31.00 K	0	0	—	++
15	M	N	241	0.20	48.00 K	0	0	—	++
8A	F	N	132	0.20	26.00 K	0	0	—	±
5E	M	N	186	0.25	3.50 D	+++	1	+	0
8B	F	N	205	0.35	25.50 K	0	0	—	±
4C	F	H.F.	114	0.50	2.00 D	++++	1	+	++++, fat
5D	M	N	145	0.50	2.30 D	++	1	+	0
8D	M	N	430	0.75	4.25 D	++	1	+	0
8C	F	N	192	0.75	3.20 to 4.20 D	++++	1	+	0
45	M	N	46	1.00	4.30 to 4.90 D	++++	1	+	0
1D	M	H.F.	80	1.00	5.00 K	0, fat	0	0	++++, fat
5B	M	N	140	1.00	3.50 D	++++	1, 2	+	0
6A	F	N	121	1.00	3.50 D	+++	1	+	0
6C	F	N	138	1.00	3.50 D	+++	1	+	0
7D	F	H.F.	77	1.00	2.00 D	+++	1	+	++, fat
7F	F	N	130	1.00	1.80 D	++	1	+	0
8E	M	N	216	1.00	3.50 D	++++	1	+	0
10A	F	N	165	1.00	3.00 D	++++	1	+	0
10B	F	N	160	1.00	3.90 D	++++	0	+	0
10C	M	N	102	1.00	3.00 D	++	1	+	0
10D	F	N	170	1.00	2.00 D	++	1	+	0

* N, normal diet; H.F., high fat-low protein diet.
† K, killed; D, died.
‡ P.T. Nec., proximal tubular necrosis (0-++++).
§ Hb.-Pos. Droplets in P.T., hemoglobin-positive droplets in proximal tubule.
‖ Liver C.L. Nec., degree of centrilobular necrosis (0-++++). Animals on high fat-low protein diet had fat as indicated.

The fat distribution was studied in frozen sections stained with Sudan III.[4] Representative sections from all three groups of animals were stained with the periodic acid-Schiff routine,[5] Lephene's benzidine stain,[6] Ralph's hemoglobin stain,[7] and an iron stain.[8]

3. Jennings, R. B.: A Simple Disposable Apparatus for Dehydration of Frozen Tissues, A. M. A. Arch. Path. **52**:195-197, 1951.

4. Lillie, R. D.: Study of Certain Oil Soluble Dyes for Use as Fat Stains, J. Tech. Methods **24**:37-42, 1944.

5. McManus, J. F. A.: The Periodic Acid Routine Applied to the Kidney, Am. J. Path. **24**:643-653, 1948.

6. Herxheimer, G.: Lepehnesche Färbung des Hämoglobins, in Handbuch der biologischen Arbeitsmethoden, edited by E. Abderhalden, Berlin, Urban & Schwarzenberg, 1924, pp. 236-240.

7. Ralph, P. H.: The Histochemical Demonstration of Hemoglobin in Blood Cells and Tissue Smears, Stain Technol. **16**:105-106, 1941.

8. Gomori, G.: Microtechnical Demonstration of Iron: A Criticism of Its Methods, Am. J. Path. **12**:655-663, 1936.

RESULTS

Group 1.—Group 1 consisted of 21 animals, 10 males and 11 females of varying ages and weights. These animals received intraperitoneal injections of carbon tetrachloride (Table 1) in doses ranging from 0.2 cc. to 1.0 cc. per 100 gm. of body

TABLE 2.—*Data on Carbon Tetrachloride Administered Subcutaneously*

Animal No.	Sex	Diet*	Weight, Gm.	CCl_4/100 Gm. Body Weight, Cc.†	Hr. Alive‡	P.T. Nec.§	Hb.-Pos. Droplets in P.T.‖	Liver C.L. Nec.¶
5	M	N	264	0.10	24.00 K	0	0	0
3	M	N	273	0.20	72.00 K	0	0	0
13	F	N	230	0.20	48.00 K	0	0	++
13A	F	N	220	0.25, PO	66.00 K	0	0	+++
1	M	N	230	0.40	72.00 K	0	0	++
2	M	N	302	0.50	31.00 K	0	0	++
7B	M	H.F.	85	0.50	53.50 K	0	0	+++
13D	F	N	190	0.50, PO	45.00 K	0	0	++
4	M	N	255	0.75	24.00 K	0	0	+
6	M	N	290	0.75	24.00 K	0	0	+
8	M	N	273	0.75	400.00 K	0	0	0
12	M	N	179	1.00	31.00 K	0	0	++
30	M	N	340	1.00	53.00 K	0	0	++
32	F	N	185	1.00	0.10 D	0	0	0
33	F	N	280	1.00	53.50 K	0	0	++
38	F	N	185	1.00	28.00 K	0	0	+++
41	F	N	212	1.00	28.00 K	0	0	+++
42	F	N	205	1.00	30.50 K	0	0	+++
43	M	N	43	1.00	48.00 K	0	0	++
44	M	N	47	1.00	48.00 K	0	0	+++
1E	M	H.F.	93	1.00	26.00 K	0, fat	0	++++, fat
2C	F	H.F.	84	1.00	29.00 K	0, fat	0	+++, fat
2D	F	H.F.	90	1.00	73.00 K	0, fat	0	+++, fat
2E	F	H.F.	89	1.00	73.00 K	0, fat	0	++++, fat
5A	M	N	129	1.00	24.00 K	0	0	+
6B	F	N	129	1.00	73.00 K	0	0	++
11A	M	N	200	1.00	1.00 K	0	0	0
11B	F	N	200	1.00	2.00 K	0	0	0
13C	F	N	210	1.00, PO	0.50 D	0	0	0
14A	F	N	210	1.00, PO	4.50 K	0	0	0
14B	F	N	220	1.00	4.50 K	0	0	0
9I	M	N	158	1.50	47.50 K	0	0	++
7	M	N	277	1.50	88.00 K	0	0	++
9J	M	N	151	1.50	47.00 K	0	0	+++
29	M	N	165	2.00	72.00 K	0	0	++
40	F	N	195	2.00	0.15 D	0	0	0
1C	M	H.F.	107	2.00	30.00 K	0, fat	0	++++, fat
2A	F	H.F.	112	2.00	96.00 K	0, fat	0	++++, fat
2B	F	H.F.	92	2.00	96.00 K	0, fat	0	++, fat
5C	M	N	146	2.00	96.00 K	0	0	+, fat
9A	F	N	165	2.00	0.15 D	0	0	0
14E	F	N	215	2.00, PO	4.25 K	0	0	0
9B	F	N	168	2.50	0.10 D	0	0	0
9C	F	N	159	3.00	0.12 D	0	0	0
9D	F	N	162	5.00	0.07 D	0	0	0

* N, normal; H.F., high fat = low protein.
† PO, paraffin oil.
‡ K, killed; D, died.
§ P.T. Nec., proximal tubular necrosis (0 - ++++).
‖ Hb.-Pos. Droplets in P.T., hemoglobin-positive droplets in proximal tubule.
¶ Liver C.L. Nec., degree of centrilobular necrosis (0 - ++++). Animals on high fat-low protein diet had fat as indicated.

weight. Fifteen of 17 animals receiving 0.35 to 1.0 cc. of carbon tetrachloride per 100 gm. of body weight died between two and five hours after the administration of the drug. Microscopic examination of the kidneys of the dead animals revealed a bilateral

necrosis of the cortical tubular epithelium. The liver was free of necrosis. Two animals, one receiving 0.35 and the other 1.0 cc. per 100 gm. of body weight, and the animals receiving less than 0.25 cc. per 100 gm. of body weight did not have the renal lesion.

The animals became lethargic about five to seven minutes after the intraperitoneal administration of 0.25 to 1.0 cc. of carbon tetrachloride per 100 gm. body weight and remained so until they died. Representative blood pressure measurements on the tail [9] showed that within the first 45 minutes the systolic blood pressure dropped precipitously from normal control levels, which were around 110 mm. Hg. to below

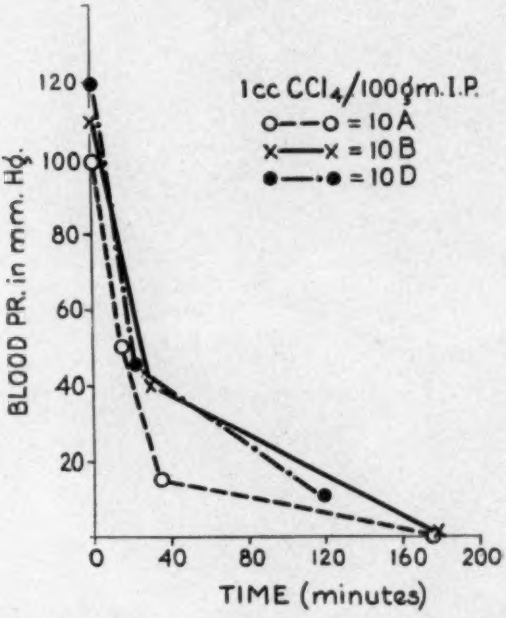

Fig. 1.—Graph of blood pressures of Rats 10A, 10B, and 10D following intraperitoneal injections of carbon tetrachloride.

70 mm. Hg (Fig. 1). Cyanosis of the ears, nose, and feet accompanied this blood pressure drop. The animals died quietly.

At postmortem examination the peritoneal cavity usually contained 3 to 6 cc. of serosanguineous fluid. The kidneys were of normal size, and the capsules stripped with ease. Surfaces made by cutting bulged, and the cut surface was usually quite pale on the cortical side of the corticomedullary junction, Zone 2 of McFarlane.[10] All other organs were grossly normal.

9. Friedman, M., and Freed, S. C.: Microphonic Manometer for Indirect Determination of Systolic Blood Pressure in the Rat, Proc. Soc. Exper. Biol. & Med. **70**:670-672, 1949.

10. (*a*) McFarlane, D.: Experimental Phosphate Nephritis in the Rat, J. Path. & Bact. **52**:17-24, 1941. (*b*) Peter, K.: Untersuchungen über Bau und Entwickelung der Niere, Jena, Gustav Fischer, 1909.

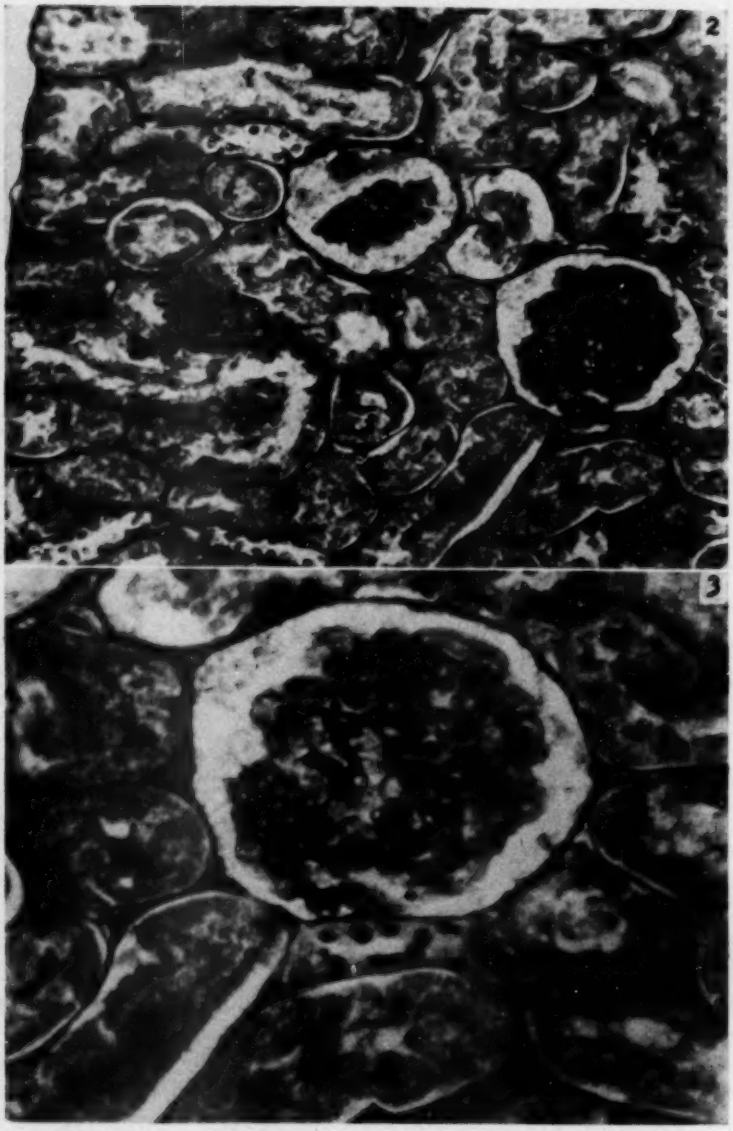

Fig. 2.—Section of kidney of Rat 8E. This animal received 1 cc. of carbon tetrachloride/100 gm. of body weight. Note necrosis of proximal tubules in the outer zone of the cortex. Periodic acid stain; × 43.

Fig. 3.—High-power view of tissue around the glomerulus of Figure 2. The glomerulus is histologically normal though hyperemic. A small amount of protein precipitate is present in Bowman's space. Note the complete necrosis of the proximal tubules in contrast to a normal distal convoluted tubule immediately beneath the glomerulus. Periodic acid stain; × 446.

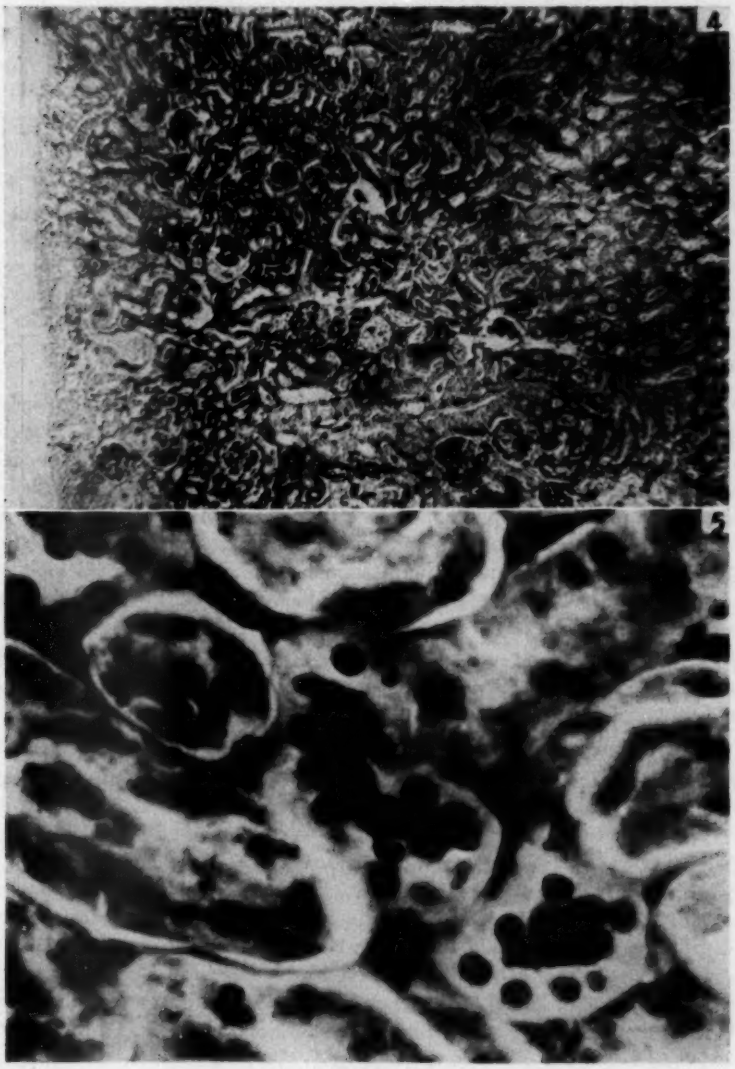

Fig. 4.—Ralph-type hemoglobin stain of kidney of Animal 8C. No counterstain has been used. Hemoglobin-positive material in the section is stained black. Zone 1 and a portion of Zone 2 of the cortex are present in the photomicrograph. Note that the largest amount of intratubular hemoglobin-positive material is in the tubules of Zone 1; × 10.

Fig. 5.—A high-power view of a similar section (Fig. 4) of kidney from Animal 8C. Hemoglobin-positive material is present in the lumen of a distal convoluted tubule. Note the small droplets of hemoglobin-positive material in the cytoplasm of the cells of the surrounding proximal tubules. Lephene stain; × 446.

Microscopic examination of the kidney revealed a cortical necrotizing nephrosis of varying severity. Many, and sometimes all, of the proximal tubules in the outermost rim of the renal cortex, Zone 1 of McFarlane,[10a] were the seat of coagulation necrosis. The nuclei were either absent or pyknotic, and beginning or advanced fragmentation of the lightly eosinophilic cytoplasm was present. The glomerular tufts were histologically normal, although Bowman's space was usually filled with large quantities of protein precipitate (Figs. 2 and 3). The distal convoluted tubules and collecting ducts were never necrotic, and the medullary portion of the kidney was histologically normal.

The brush border of the intact proximal tubular cells with normal or pyknotic nuclei stained positively with periodic acid, as did the brush border in the control animals. With cell death and nuclear fragmentation, the bright reddish purple of this stain disappeared from the brush border. The lumens of many of the tubules were filled with protein precipitate, which was brightly eosinophilic, stained positively with periodic acid,[8] and frequently the hemoglobin stain was also positive. Many cells of the proximal tubules contained variable numbers of brightly eosinophilic hyaline droplets which were periodic acid- and hemoglobin-positive. The brush border was also occasionally hemoglobin-positive, especially when the underlying cells contained droplets or when the adjacent tubular lumen was filled with hemoglobin-positive protein precipitate. Upon cell death, with disappearance of nuclei and smudging of the cytoplasm of the proximal tubular cells, these droplets disappeared. The droplets did not contain stainable iron. Rarely, small globules of cytoplasmic neutral fat were present in some of the injured cells between the nucleus and the brush border.

No significant necrosis of the liver[11] was discovered in rats with necrotizing nephrosis, but centrilobular necrosis was apparent in the three animals of this series which were killed after 24 hours.

Sections of lung, bowel, heart, gonad, spleen, and adrenal gland were normal.

Group 2.—Group 2 consisted of 45 rats which received subcutaneous injections of carbon tetrachloride. From Table 2 it will be seen that no instances of tubular necrosis occurred in these animals, 21 of which were male and 24 female. These rats weighed between 43 and 340 gm. and were given carbon tetrachloride in one or both flanks in doses ranging from 0.1 to 5.0 cc. of carbon tetrachloride per 100 gm. of body weight. Animals 13A, 13C, 13D, 14A, and 14E received subcutaneous administration of carbon tetrachloride diluted 1 : 1 with liquid petrolatum. All others received undiluted carbon tetrachloride. All animals receiving 1.5 cc. of carbon tetrachloride per 100 gm. or less of body weight lived and were killed at varying intervals by a blow to the head. The animals became lethargic when doses of 1.0 to 1.5 cc. of carbon tetrachloride per 100 gm. of body weight were administered, and doses of 2.0 cc. of carbon tetrachloride per 100 gm. or larger frequently were lethal within 10 minutes.

There were no notable gross lesions except in the liver, which was enlarged and mottled diffusely dark red and brown.

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11. Stowell, R. E., and Lee, C. S.: Histochemical Studies of Mouse Liver After Single Feeding of Carbon Tetrachloride, Arch. Path. **50**:519-537, 1950. Williams, W. L., and Gregory, M. A.: Effects of Carbon Tetrachloride and of Starvation on Cytoplasmic Histochemistry of Parenchymal Cells of Mouse Livers, Fed. Proc. **10**:374, 1951.

Microscopic examination of the kidney showed no necrosis in the proximal convoluted tubules, although cloudy swelling was usually present. Small amounts of protein precipitate were occasionally seen in Bowman's space, but this may be normal, since most species of rats ordinarily have a measurable proteinuria.[12] The lumens of the proximal tubules never contained hemoglobin-positive material. The tubular epithelium never contained the large eosinophilic hemoglobin-positive droplets seen in Group 1. The medulla of the kidney was histologically normal.

Microscopic examination of the liver showed centrilobular foci of necrosis or fat infiltration, depending upon the number of hours after the injection. These changes were most marked at 48 to 72 hours.

Sections of lung, spleen, bowel, gonad, and adrenal gland were not notable.

Group 3.—Group 3 consisted of 26 animals which were not experimented upon. They were used as controls for organ size and staining characteristics. Twelve animals in this group were on a high fat-low protein diet and had diffuse fatty livers and sudanophilic material in the proximal and distal tubular cells of the kidney.

COMMENT

These experiments have shown that the rat kidney may react in a specific fashion to the acute administration of carbon tetrachloride intraperitoneally in doses above 0.5 cc. per 100 gm. of body weight. This dose or greater was almost invariably fatal, and the animal rarely survived more than five hours. The most striking finding at postmortem examination was necrosis of the proximal tubules in the outer strip of the cortex.

Many investigators have studied the effect of carbon tetrachloride on rats. Most of these experiments, however, have been concerned with the hepatotoxic rather than the nephrotoxic action of this chlorinated hydrocarbon. It has been administered subcutaneously, orally, intraperitoneally, and by inhalation. No renal lesion of the type observed to follow intraperitoneal administration of carbon tetrachloride in our experiments has been reported. Usually only small quantities of carbon tetrachloride have been used.

György, Seifter, Tomarelli, and Goldblatt [13] noted necrotizing cortical nephrosis in some of the rats that died during the course of experiments in which the effects of "chronic inhalation" of carbon tetrachloride (300 ppm, seven hours a day, five days a week) were compared in animals on different diets. They found that male rats on a high fat-low protein diet were very susceptible, dying in less than 150 days, with many necrotic tubules in the renal cortex. Methionine protected them against the lesion, and female rats were less susceptible than males. In our experiments 11 rats and 12 controls were placed for 24 days on the same high fat-low protein diet.[13] These animals (see H. F. in Tables 1 and 2) had fatty livers and fat in the proximal tubules of the kidneys but were no more susceptible to the acute administration of large doses of carbon tetrachloride than animals on a normal diet.

12. Gilson, S. B.: Studies on Proteinuria in the Rat, Proc. Soc. Exper. Biol. & Med. **72**:608-613, 1949.

13. György, P.; Seifter, J.; Tomarelli, R. M., and Goldblatt, H.: Influence of Dietary Factors and Sex on the Toxicity of Carbon Tetrachloride in Rats, J. Exper. Med. **83**:449-462, 1946.

Acute and chronic carbon tetrachloride experiments have also been carried out in other experimental animals, notably dogs, cats, mice, and rabbits.

Gardner, Grove, Gustafson, Mauri, Thompson, Wells, and Lamson [14] found no kidney lesions in either puppies or adult dogs given carbon tetrachloride orally, intravenously, rectally, and intraperitoneally. Chandler and Chopra [15] studied the effects of oral carbon tetrachloride administration in cats and stated that only 4 out of 49 cats survived doses of 0.5 cc. of carbon tetrachloride per kilogram of body weight. They observed extensive fatty degeneration and frequently necrosis of the proximal convoluted tubules. However, Hall and Shillinger [16] gave similar oral doses to kittens and found the compound to be nonlethal. Smyth [17] did a series of long-term inhalation experiments on rats, guinea pigs, and monkeys in order to determine the minimum toxic concentration of carbon tetrachloride in air. Marked necrosis of the proximal tubules was not observed.

Mauro [18] performed inhalation experiments in rabbits but made no mention of necrosis of proximal tubular cells. Oliver,[19] however, observed necrosis of the tubular cells of the terminal portion of the proximal convoluted tubule in rabbits given two consecutive doses subcutaneously of 4 cc. of carbon tetrachloride. This lesion is similar to that seen in the rats given carbon tetrachloride intraperitoneally in our experiments. Opie [20] reported the absence of tubular necrosis in the kidneys of rats given single subcutaneous injections of carbon tetrachloride, which is the same as our finding.

Studies using chloroform, $CHCl_3$, which is a chlorinated hydrocarbon closely related to carbon tetrachloride, confuse the picture further because chloroform diluted with liquid petrolatum and given orally and subcutaneously produces proximal tubular necrosis. Eschenbrenner and Miller [21] have demonstrated necrosis in the kidneys of male mice which lived 24 hours after the oral administration of chloroform. Opie and the present investigators have both observed a similar lesion in rats given subcutaneous administration of chloroform in liquid petrolatum (Fig. 7). Carbon tetrachloride in liquid petrolatum given subcutaneously is not nephrotoxic. No adequate explanation is available to account for failure of the lesion to develop in rats given

14. Gardner, G. H.; Grove, R. C.; Gustafson, R. K.; Mauri, E. D.; Thompson, M. J.; Wells, H. S., and Lamson, P. D.: Studies on the Pathological Histology of Experimental Carbon Tetrachloride Poisoning, Bull. Johns Hopkins Hosp. **36**:107-133, 1925.

15. Chandler, A. C., and Chopra, R. N.: The Toxicity of Carbon Tetrachloride to Cats: A Warning, Indian M. Gaz. **60**:406-407, 1925.

16. Hall, M. C., and Shillinger, J. E.: Miscellaneous Tests of Carbon Tetrachloride as an Anthelmintic, J. Agric. Res. **23**:163, 1923.

17. Smyth, H. F.; Smyth, H. F., Jr., and Carpenter, C. P.: The Chronic Toxicity of Carbon Tetrachloride: Animal Exposures and Field Studies, J. Indust. Hyg. & Toxicol. **18**:277-298, 1936.

18. Mauro, G.: Intossicazione da tetracloruro di carbonio, Clin. med. ital. **61**:192-201, 1930; cited by Smetana.[16]

19. Oliver, J.; MacDowell, M., and Tracy, A.: The Pathogenesis of Acute Renal Failure Associated with Traumatic and Toxic Injury: Renal Ischemia, Nephrotoxic Damage and the Ischemuric Episode, J. Clin. Invest. **30**:1307-1438, 1951.

20. Opie, E. L.: The Effect of Injury by Toxic Agents upon Osmotic Pressure Maintained by Cells of Liver and of Kidney, J. Exper. Med. **91**:285-294, 1950.

21. Eschenbrenner, A. B., and Miller, E.: Sex Differences in Kidney Morphology and Chloroform Necrosis, Science **102**:302-303, 1945.

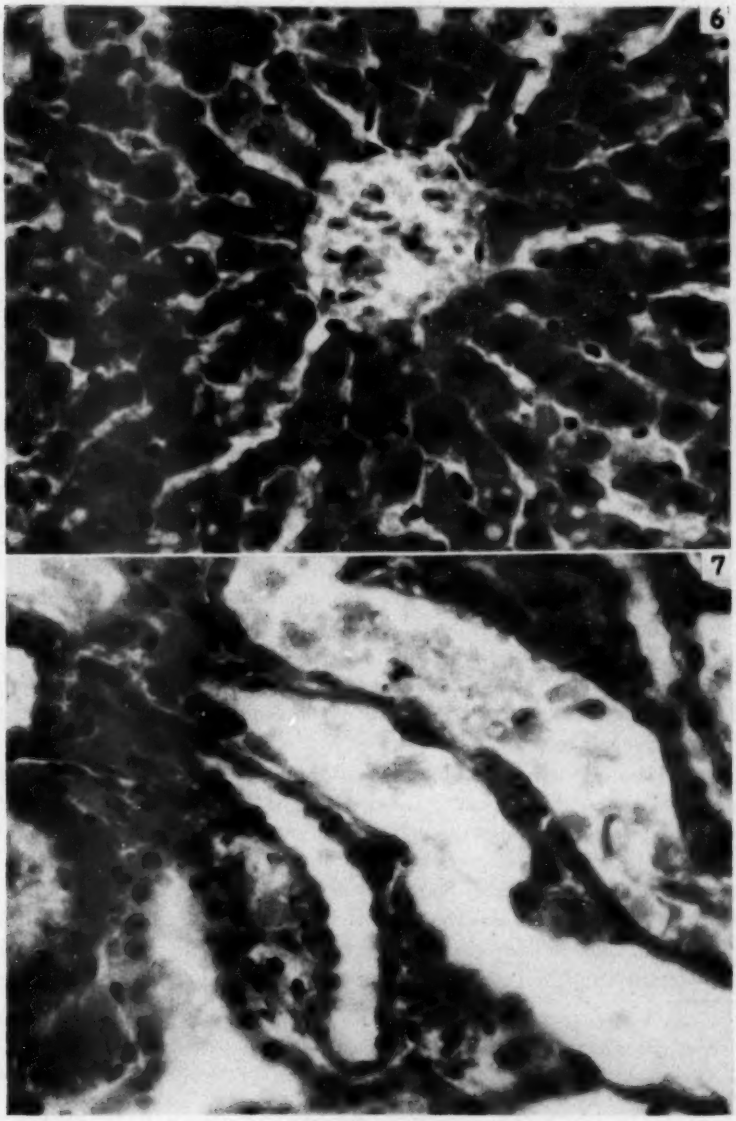

Fig. 6.—Section of liver from Animal 8E. A central vein with surrounding intact parenchymal tissue is present in the center of the photomicrograph. This animal died three and one-half hours after injection of 1 cc. of carbon tetrachloride/100 gm. of body weight. Hematoxylin and eosin; × 43.

Fig. 7.—Section of kidney from Animal 12A. This female rat weighed 200 gm. and received subcutaneously 0.25 cc. of chloroform in liquid petrolatum/100 gm. of body weight. It was killed 72 hours later. Necrosis of proximal tubular cells in Zone 1 was noted. Regeneration beneath the dead cells is shown clearly in this photomicrograph. Three mitotic figures in the regenerating epithelium are present in the lower right-hand corner of the section. Hematoxylin and eosin; × 446.

carbon tetrachloride subcutaneously. It is possible that carbon tetrachloride, when administered by this route, is absorbed more slowly than would be expected because of pooling of carbon tetrachloride, an excellent fat solvent, in subcutaneous fat.

A shunt phenomenon, such as that described by Trueta and co-workers [22] in rabbits in shock, may be in operation, for our animals were in deep shock for at least an hour before death (Fig. 1). It has been shown that the blood supply to the kidney is greatly reduced in shock, and anoxia on this basis would probably first injure the highly differentiated cells of the proximal tubule.[23] Oliver [19] has recently graphically illustrated a patchy ischemia in the renal cortex of experimental animals which had received various toxic agents.

The possibility of a direct toxic action of carbon tetrachloride on the proximal tubular cells is more probable, since carbon tetrachloride given intraperitoneally (rapid absorption with shock) and chloroform given subcutaneously (slow absorption without shock) produce an identical lesion.

The origin and significance of the hemoglobin-positive droplets in the cytoplasm of the renal tubular cells are unknown. They appear identical with similar intracellular droplets described in rats with hemoglobinuria produced by intraperitoneal injection of hemoglobin solutions.[24] If it is permissible to assume that the Lephene and Ralph benzidine-H_2O_2 type hemoglobin stains are specific for this substance, then these droplets probably represent small quantities of this reabsorbed protein. Similar intraluminal and intracellular hemoglobin-positive material has been observed in the kidneys of dogs approximately five hours after subcutaneous administration of Clostridium perfringens toxin.[25] This toxin is a well-known hemolytic agent and might conceivably cause increased hemoglobin excretion, with resultant tubular reabsorption. A similar sequence of events is perhaps occurring in rats given carbon tetrachloride intraperitoneally.

The renal lesion seen in our animals was not similar to the so-called "lower nephron nephrosis" described by Lucke [1a]; Dunn, Gillespie, and Niven [1b]; Mallory,[1c] and Woods [1d] or to the changes in the upper nephron seen by Smetana,[1e] Moon,[1f] and McManus and Rutledge [1g] in cases of human intoxication with carbon tetrachloride. Human intoxication with carbon tetrachloride usually takes place through ingestion or inhalation, and in fatal cases the patients frequently do not die for 5 to 10 days. At this time the most striking changes are present in the lower nephron, and a histological diagnosis of "lower nephron nephrosis" is made. Our experiments and the

22. Trueta, J.; Barclay, A. E.; Franklin, K. J.; Daniel, P. M., and Pritchard, M. L.: Studies of the Renal Circulation, Springfield, Ill., Charles C Thomas, Publisher, 1947.

23. Van Slyke, D. D.; Phillips, R. A.; Hamilton, P. B.; Archibald, R. M.; Dole, V. P., and Emerson, K., Jr.: Effect of Shock on the Kidney, Tr. A. Am. Physicians **58**:119-128, 1944. Phillips, R. A.; Dole, V. P.; Hamilton, P. B.; Kendall, E.; Archibald, R. M., and Van Slyke, D. D.: Effects of Acute Hemorrhagic and Traumatic Shock on Renal Function of Dogs, Am. J. Physiol. **145**:314-336, 1946.

24. Oliver, J.: Renal Function, New York, Josiah Macy, Jr., Foundation, 1949. Lippman, R. W.; Ureen, H. J., and Oliver, J.: Mechanism of Proteinuria: III. A Comparison of Functional and Structural Aspects of the Effects of Certain Intraperitoneally Administered Proteins on Hemoglobin Excretion in the Rat, J. Exper. Med. **93**:325-336, 1951.

25. Berg, M.; Levinson, S. A., and Wang, K. J.: Effect of Experimental Shock Induced by Clostridium Perfringens Toxins on the Kidneys of Dogs, A. M. A. Arch. Path. **51**:137-153, 1951.

recent work of Oliver [19] suggest that the early lesion of human carbon tetrachloride intoxication may possibly be in the proximal tubules of the kidney and that the rather nonspecific histological picture of lower nephron nephrosis is a late manifestation.

SUMMARY

The intraperitoneal administration of carbon tetrachloride to rats on a normal diet was lethal in less than five hours when the dose was greater than 0.5 cc. of carbon tetrachloride per 100 gm. of body weight and was almost invariably accompanied by necrosis of the proximal tubules in the outermost zone of the renal cortex.

Age, sex, and a high fat-low protein diet did not influence the production of the renal lesion.

The pattern of the necrosis did not follow that of "lower nephron nephrosis." The mechanism of production of the lesion is unknown. Either shock, with decreased renal blood flow, or a direct toxic action are considered the most likely cause of the injury and are being further investigated.

HISTOCHEMICAL STUDIES IN CYTOMEGALIC INCLUSION DISEASE

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THE FACT that both the intranuclear inclusions of cytomegalic inclusion disease [1] and the hematoxylin-staining bodies of disseminated lupus erythematosus assume a peculiar reddish violet color in hematoxylin and eosin-stained sections suggested that changes of a similar nature might be present in these two structures. Klemperer and co-workers [2] have concluded from their results obtained by photometric cytochemical methods that the hematoxylin-staining bodies of lupus erythematosus contained partially depolymerized desoxyribonucleic acid. Similar studies were therefore undertaken on the inclusion bodies.

MATERIALS AND METHODS

The material for study was obtained at autopsy 16 hours post mortem from a 48-day-old prematurely born infant who died of generalized cytomegalic inclusion disease. Additional material was obtained by routine excision of the parotid and submaxillary salivary glands of infants at autopsy. Two salivary glands (excised 27 hours post mortem) containing cytomegolic inclusions were found. Tissues were fixed in 20% formalin and in Carnoy's acetic alcohol (1:3).

Among the stains used to help characterize the inclusions were hematoxylin and eosin, methyl green, Feulgen (controlled), periodic acid-Schiff, Millon (controlled with and without addition of nitrite), and toluidine blue O. Because of previous dissapointing experiences with desoxyribonuclease on paraffin sections, no such studies were undertaken.

Photometric studies in the visible spectrum were done on methyl green- and Feulgen-stained Carnoy-fixed sections according to the method described by Pollister and Ris [3] and Pollister and Leuchtenberger.[4] The apparatus used was similar to that described by Pollister and Moses.[5] The methyl green-stained sections were measured with a Wratten 26 filter at 625 mμ and the Feulgen-stained sections with a Farrand interference filter at 559 mμ.

In the ultraviolet region, unstained formalin-fixed sections were measured in Dr. B. Gueft's laboratory on a similar apparatus employing a Beckmann spectrophotometer adapted as a monochromator and using a hydrogen lamp as light source. A reflecting objective (N. A. 0.72) was used, along with a quartz ocular and condenser in the microscope. A 1P28 photomultiplier

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1. Wyatt, J. P., and others: J. Pediat. **36**:271, 1950.
2. Klemperer, P., and others. Arch. Path. **49**:503, 1950.
3. Pollister, A. W., and Ris, H. R.: Cold Spring Harbor Symp., Quant. Biol. **12**:147, 1947.
4. Pollister, A. W., and Leuchtenberger, C.: Proc. Nat. Acad. Sc. **35**:111, 1949.
5. Pollister, A. W., and Moses, M. J.: J. Gen. Physiol. **32**:567, 1949.

tube with a D. C. amplifier and galvanometer was used as the photometric part of the apparatus. A more complete description of the ultraviolet spectrophotometric apparatus that was used is given by Gueft and Laufer.[6]

RESULTS

The intranuclear inclusions appeared more refractile than normal nuclei in unstained sections. They were colored uniformly reddish violet with hematoxylin and eosin, in contrast to the marginated nuclear chromatin, which appeared blue. They also stained with the Feulgen reaction, methyl green, and the Millon reagent. This last reagent colored the inclusions a darker orange-brown than it did the surrounding nuclear rim and cytoplasm of other cells. Toluidine blue O stained the inclusions blue or greenish blue, and the periodic acid-Schiff technique left them colorless.

The intracytoplasmic inclusions stained like the intranuclear inclusions, with the exception that they were colored with the periodic acid-Schiff reagent even

Methyl Green and Feulgen Extinction Values of Intranuclear Inclusion Bodies and of Normal Duct Cell Epithelial Nuclei in Salivary Glands

Inclusion Body No.	Methyl Green Extinction Values*	Feulgen Extinction Values†	Ratio Feulgen to Methyl Green	Nucleus No.	Methyl Green Extinction Values*	Feulgen Extinction Values†	Ratio Feulgen to Methyl Green
1.....	0.125	0.529	4.23	1.....	0.285	0.423	1.49
2.....	0.069	0.484	7.06	2.....	0.229	0.429	1.87
3.....	0.092	0.499	5.43	3.....	0.246	0.452	1.84
4.....	0.049	0.493	10.07	4.....	0.196	0.456	2.86
5.....	0.083	0.653	7.87	5.....	0.168	0.452	2.69
6.....	0.052	0.566	10.89	6.....	0.197	0.477	2.42
7.....	0.115	0.571	4.97	7.....	0.267	0.374	1.40
8.....	0.275	0.637	2.32	8.....	0.204	0.374	1.44
9.....	0.072	0.456	6.34	9.....	0.216	0.311	1.44
10.....	0.058	0.268	4.62	10.....	0.150	0.311	2.07
11.....	0.121	0.393	3.24	11.....	0.168	0.324	1.93
12.....	0.113	0.343	3.03	12.....	0.129	0.229	1.91
13.....	0.088	0.294	3.35	13.....	0.242	0.301	1.24
14.....	0.069	0.315	4.55				
15.....	0.063	0.357	5.65				
16.....	0.014	0.277	20.50				
Mean.....	0.091	0.446	6.51		0.207	0.378	1.85

* Measured at 625 mμ.
† Measured at 559 mμ.

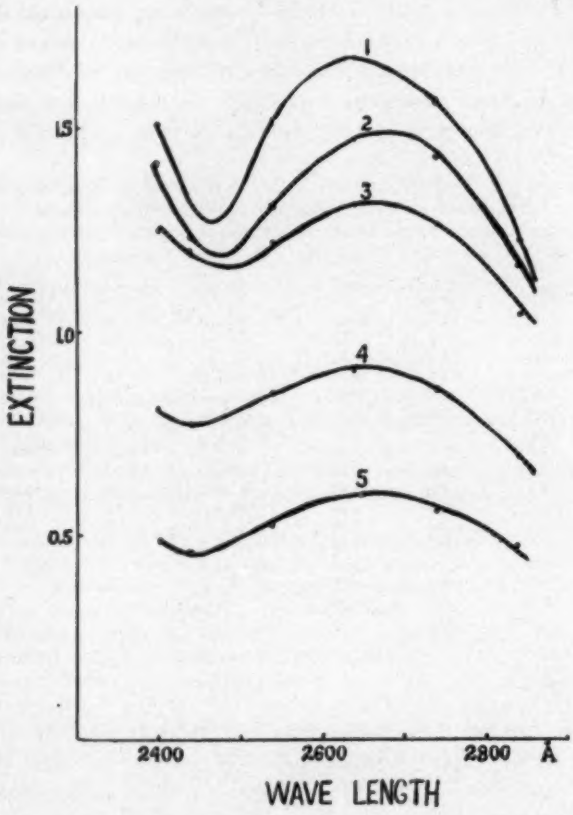
after antecedent saliva digestion and exhibited slight but definite metachromasia with toluidine blue O. This metachromasia was more intense than that given by normal nuclei and was found in the cytomegalic cells of nonsecreting organs, as well as in the salivary glands.

Microphotometric studies were carried out on well-defined inclusion bodies and normal nuclei with use of the Feulgen and methyl green stains. Sections of salivary glands containing inclusions were stained with methyl green according to the modification of Pollister and Leuchtenberger,[4] and the extinction of individual inclusion bodies and normal nuclei were measured with red light at 625 mμ. The same section was then stained by the Feulgen technique, in which the preliminary acid hydrolysis also decolorizes the methyl green. The extinction of the identical inclusion bodies and normal nuclei were then measured with green light at 559 mμ.

6. Gueft, B., and Laufer, A.: Personal communication to the authors.

The ratio of the extinction value of the Feulgen to that of the methyl green was then computed for each inclusion body and normal nucleus measured. The values are recorded in the Table.

It will be noted that (1) in all but one instance the ratio for the inclusion bodies was distinctly higher than that for any of the normal nuclei, (2) the range for the inclusion bodies (2.32 to 20.5) was much wider than that for the normal nuclei (1.24 to 2.69), (3) although the averages of the Feulgen extinctions for normal



Ultraviolet extinction curves for inclusion bodies (*1*, *2*, and *3*) and for normal nuclei (*4* and *5*) in kidney tubules of infant with cytomegalic inclusion disease.

nuclei and inclusion bodies were fairly close, those of the methyl green extinctions were distinctly lower for the inclusion bodies. The last finding excludes the possibility that the error introduced by nonhomogeneity of the normal nucleus compared with that of the inclusion is sufficient to account for the differences observed.

Absorption curves for three intranuclear inclusions and two normal nuclei of the kidney tubule in the ultraviolet region of the spectrum are plotted in the Chart. All curves show a peak absorption between 2,600 and 2,700 A.

COMMENT AND CONCLUSIONS

The ultraviolet absorption maxima between 2,600 and 2,700 A are due to the pyrimidine portion of the molecule of either ribonucleic or desoxyribonucleic acid. The finding of similar absorption peaks in all the curves indicates the presence of one or both of these nucleic acids in the inclusions. The difference in level between the curves for the inclusions and those for the normal nuclei suggests a higher nucleic acid concentration in the former.

The fact that some of this nucleic acid is desoxyribonucleic acid is clearly demonstrated by the positive Feulgen and methyl green stains. The peculiarity of this nucleic acid, however, is brought out by the Feulgen-methyl green extinction ratios. Pollister and Leuchtenberger [4] and Kurnick [7] have pointed out that the methyl green stain may be used as an indicator of the state of polymerization of desoxyribonucleic acid. With increased depolymerization the methyl green stainability decreases. Since the Feulgen reaction is unaffected by these subtle changes in physical state, the ratio of the intensity of the Feulgen stain to that of the methyl green can be used as an index of depolymerization. The results given in the Table demonstrate (1) that the intranuclear inclusions contain desoxyribonucleic acid which has undergone depolymerization, (2) that the degree of depolymerization is variable for different inclusions. The variability is not unexpected, since the inclusions age with their host cells and may eventually undergo lysis.

The high refractility of the inclusions plus their tendency toward greater Millon positivity indicates a high protein content.

The discrepancies in staining characteristics between the two types of inclusion bodies, as revealed by the periodic acid-Schiff technique and toluidine blue O, suggest the presence of a carbohydrate-containing fraction that is either adsorbed or incorporated into the intracytoplasmic inclusions and absent in the intranuclear inclusions.

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7. Kurnick, N. B.: J. Gen. Physiol. **33**:243, 1950.

RHABDOMYOSARCOMA OF THE VULVA AND VAGINA

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DURING the course of a recent survey [1] of sarcoma of the female genital tract, based on material filed at the Armed Forces Institute of Pathology, seven nonepithelial tumors of the vulvovaginal region were studied; four of these were examples of sarcoma botryoides; one was a uniform granular cell myoblastoma, and two were classified as rhabdomyosarcoma. These last two cases furnish the basis for the present report. Although sarcoma botryoides of the vulva and vagina is infrequent, tumors composed principally of muscle cells are even less frequently encountered in this area; however, a few scattered neoplastic muscle cells are almost constantly present in botryoid sarcomas. McFarland,[2] in 1935, reviewed the literature relevant to dysontogenetic tumors of the urogenital ridge derivatives and found 165 cases of vulvar vaginal sarcomas. Seventy-five, the large majority occurring in infants, were labeled botryoid, or grape-like, by the original observer. In only five of these had neoplastic muscle cells been plentiful enough to warrant a diagnosis of rhabdomyoma or rabdomyosarcoma. The remaining 80 cases comprised a variety of sarcomas, in many of which documentation or microscopic description was inadequate for a diagnosis more specific than "sarcoma."

Of the five cases labeled rhabdomyoma or rhabdomyosarcoma in McFarland's review, those of Brumwell,[3] Lockwood,[4] and Gurd [5] were botryoid sarcomas in infants, apparently with a fairly high proportion of so-called rhabdomyoblasts. Kaschewarowa-Rudnewa [6] reported two cases, one, a classic rhabdomyosarcoma arising from the anterior vaginal wall of a 15-year-old girl, the other, a similar

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1. Grady, H. G., and Ober, W. B.: Unpublished data.

2. McFarland, J.: Dysontogenetic and Mixed Tumors of the Urogenital Region, with Report of New Case of Sarcoma Botryoides Vaginæ in Child, and Comments upon Probable Nature of Sarcoma, Surg., Gynec. & Obst. **61**:42-57, 1935.

3. Brumwell, J.: Rhabdomyosarcoma of the Uterus and Vagina in an Infant, Newcastle M. J. **10**:157-161, 1930.

4. Lockwood, C. D.: Rhabdomyosarcoma of the Vulvar Orifice in Children: Report of Case, Arch. Surg. **14**:860-867, 1927.

5. Miller, C. J., and Gurd, F. B.: Malignant Rhabdomyoma of the Vagina in Children, Surg., Gynec. & Obst. **11**:391-397, 1910.

6. Kaschewarowa-Rudnewa, W.: Beitrag zur pathologischen Anatomie der weiblichen Scheide bei Menschen und Tieren, Arch. path. Anat. **54**:63-77, 1872.

tumor arising in the uterus of a female dog. Since McFarland's study, a number of single cases of sarcoma botryoides and four other vulvovaginal sarcomas have been reported. Keller [7] described an example of fibrosarcoma arising in the labium majus of a 58-year-old woman, and Hauptman and Taussig [8] reported a fibrosarcoma arising in the labium majus of a 7-month-old infant who died one year after operation. In neither of these cases were any neoplastic muscle cells identified. Bernstein [9] reported a case of vulvar sarcoma in a 45-year-old woman. The tumor was classified as a "polymorphous cell sarcoma," but no mention was made of any muscular component.

Sadler and Dockerty [10] reported a rather unusual vaginal tumor in a 40-year-old woman and classified it as a malignant myoblastoma of the vulva. They considered it to be the malignant variant of the granular cell myoblastoma of Abrikossoff. The tumor was composed of irregular and spindle-shaped cells with intensely eosinophilic cytoplasm. Some of the tumor cells exhibited granularity in their cytoplasm, but cross striations were not demonstrable. In some respects the tumor they described resembles those in the two cases that we are reporting. To classify this tumor as the malignant variant of the granular cell myoblastoma raises the question of the nature of its component cells, as well as the nature of myoblasts and their role in its formation. While in the case reported by Sadler and Dockerty the growth is clearly a malignant tumor of muscle cells, its relationship to the granular cell myoblastoma is a matter of interpretation.

In 1946, Arthur Purdy Stout [11] reviewed the problem of rhabdomyosarcoma, collating 107 cases from the literature and adding 14 cases from his own material. This survey was limited to tumors of the soft tissues and did not include tumors arising in the female genital tract.

If rhabdomyosarcoma is distingushed from sarcoma botryoides and if our enumeration of published cases is complete, only two cases of rhabdomyosarcoma of the vulvovaginal area have been reported previously. Rhabdomyosarcoma of the uterus is somewhat more frequent. Campbell,[12] in 1940, presented a case of his own and surveyed those previously reported, accepting 21 as satisfactory examples of the uterine tumor. Kulka and Douglas,[13] in 1952, in contributing a case from their own experience, again reviewed the previously reported cases and accepted 19, only 1 of which had been reported during the 12 years elapsing since Campbell's survey. That only 13 cases were accepted by both reviewers indicates the order of magnitude of agreement and the value of enumerating previously reported cases of this or that rare tumor.

CASE 1 (A.F.I.P. Acc. 150253).—A 68-year-old white married woman was admitted because of vaginal bleeding and a foul vaginal discharge of three weeks' duration. She had passed through the menopause 20 years previously. There had been no bleeding or discharge until the present

7. Keller, J.: Fibrosarcoma of Labium Vulvae, Canad. M. A. J. **64**:534-536, 1951.

8. Hauptman, H., and Taussig, F. J.: Sarcoma of the Vulva in a 7-Month-Old Infant, J. Pediat. **16**:350-355, 1940.

9. Bernstein, P.: Sarcoma of the Vulva, Am. J. Surg. **45**:591-593, 1939.

10. Sadler, W. P., and Dockerty, M. B.: Malignant Myoblastoma Vulvae, Am. J. Obst. & Gynec. **61**:1047-1055, 1951.

11. Stout, A. P.: Rhabdomyosarcoma of the Skeletal Muscles, Ann. Surg. **123**:447-472, 1946.

12. Campbell, C. M.: Rhabdomyosarcoma of the Corpus Uteri, Arch. Path. **30**:607-613, 1940.

13. Kulka, E. W., and Douglas, G. W.: Rhabdomyosarcoma of the Corpus Uteri: Report of Case, Associated with Adenocarcinoma of the Cervix, with Review of Literature, Cancer **5**:727-736, 1952.

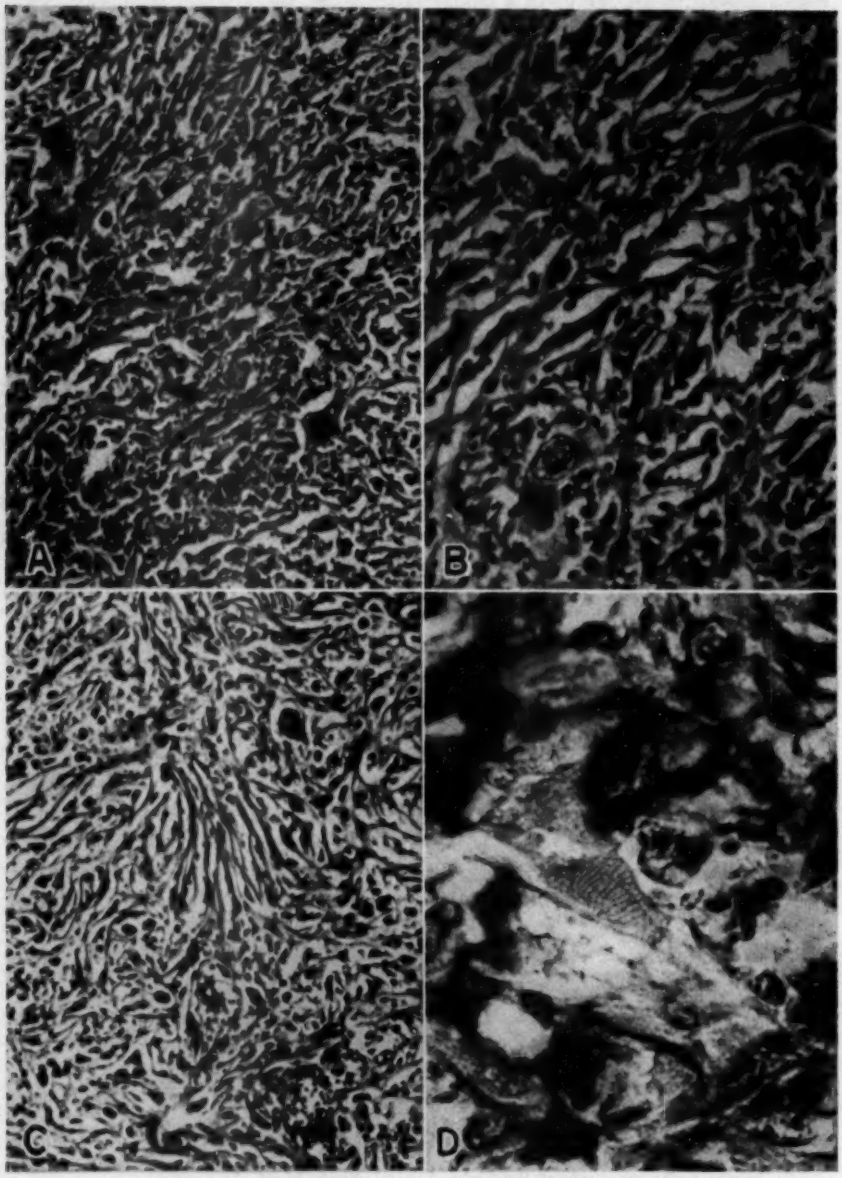

Fig. 1.—Vaginal rhabdomyosarcoma. *A*, general pattern of tumor; × 125. *B*, bizarre cells among "strap" cells with abundant eosinophilic cytoplasm; × 275. *C*, anastomosing and ramifying tumor cells. Iron hematoxylin; × 150. *D*, transverse striations (retouched). Masson trichrome; × 1000.

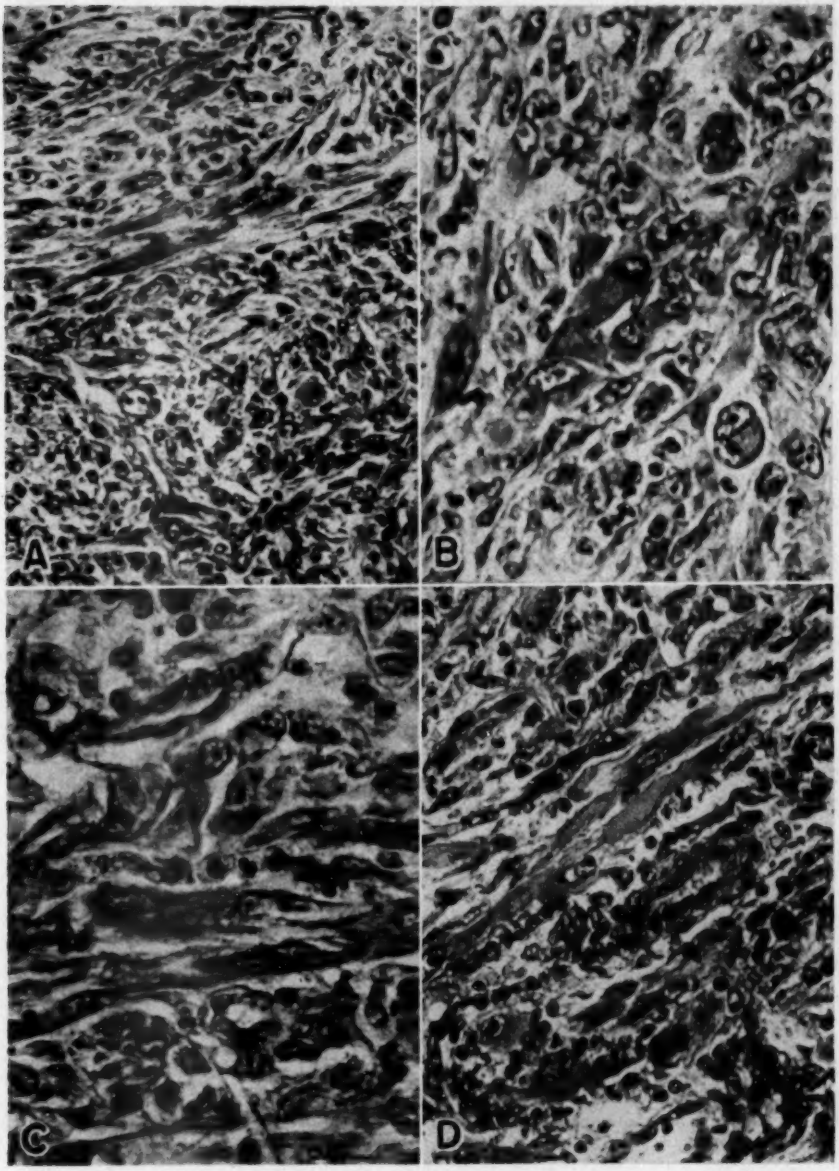

Fig. 2.—Vulvar rhabdomyosarcoma. *A*, general pattern of tumor; × 275. *B*, bizarre cells, binucleate forms; × 500. *C*, nuclear structure, tandem arrangement, formation of syncytia. Iron hematoxylin; × 700. *D*, transverse striations (retouched). Masson trichrome; × 400.

illness. Three years previously right radical mastectomy had been performed for carcinoma of the breast. Physical examination revealed a marital introitus with adequate support. A polypoid friable rounded mass, 4 cm. in diameter, arose from the anterior vaginal wall anterior to the cervix. The tumor was fairly mobile and was attached to the vaginal mucosa by a broad base but was not fixed to underlying structures. The uterus was not palpable. No masses could be felt in the adnexal regions. A biopsy was performed. Eighteen days later a complete hysterectomy, bilateral salpingo-oophorectomy, and subtotal vaginectomy were performed. No follow-up data are available.

Gross Appearance.—The uterus measured 6 by 1.8 by 3 cm. The serosa was smooth and glistening. The myometrium was thin and flabby. The endometrium was thin and smooth. The cervix was small and exhibited a few small Nabothian cysts. The Fallopian tubes and ovaries were atrophic and not remarkable. Arising from the anterior vaginal wall was a polypoid mass of firm tissue measuring 3.8 by 3 by 1.9 cm. It was attached by a moderately broad base to the vaginal mucosa and appeared to arise from the tissue subjacent to the mucosa. Apparently exophytic, it did not penetrate through the vaginal wall. The surface of the polypoid tumor was hemorrhagic and somewhat granular; the cut surface was firm, homogeneous, and grayish pink, with occasional yellowish flecks and a few small areas of recent hemorrhage.

Microscopic Description.—No significant lesions but only the changes of senile involution were found in the uterus, tubes, or ovaries. Chronic cystic cervicitis was of moderate severity. On microscopic examination of hematoxylin- and eosin-stained sections the cells comprising the tumor had substantially no organized pattern, except for a slight tendency toward fasciculation and whorling. The tumor was composed of large spindle cells which varied greatly in size and shape. Many of the cells were fusiform, but about an equal number were broad and strap-like; some cells were racquet-shaped or tadpole-shaped; still others were irregularly rounded. All these cell forms exhibited an intensely eosinophilic cytoplasm and large vesicular nuclei with both fine and coarsely clumped chromatin; in many nuclei a single prominent nucleolus was visible, usually basophilic, but occasionally acidophilic. The eosinophilic cytoplasm was rather refractile; under reduced illumination it was often longitudinally fibrillar and occasionally granular. The granularity was conspicuous in cells undergoing degenerative changes and with pyknotic nuclei, but it was rare and focal in cells without apparent degenerative changes. Many irregular multinucleated tumor giant cells were seen. Some of these were strap-like, with their nuclei arranged in tandem formation; others were in the form of very large, irregularly branching syncytial masses. Any number of transitional forms, from the simple tumor cells to the more complex forms, could be identified. Mitotic figures were abundant, and bizarre mitoses were numerous. In addition, occasional cells were smaller and seemed to have less cytoplasm; it was not possible to determine whether these represented artifacts in the form of portions of cells not in the section or whether they were less well-developed cells of the same type as the commoner components. Hemorrhage and necrosis were not prominent features, but individual cells exhibited severe degenerative changes. Practically no intercellular substance was visible in hematoxylin and eosin-stained sections. Most of the tumor cells were clearly bipolar; their lateral cell margins were invaribly distinct, but their poles, often in apposition to other cells, were quite indistinct, a further suggestion of syncytium formation.

Sections prepared by Masson's trichrome technique and with Gomori's trichrome stain failed to demonstrate any collagenous matrix; no intercellular substance was seen. The cytoplasm of the tumor cells was stained bright reddish orange by both procedures. Sections treated with silver failed to reveal any reticulum fibers in association with the tumor cells. Most of the tumor cells exhibited

argentaffin properties, but the silver seemed to precipitate on the surface of the cells, giving them a tubular appearance. In all of these preparations the longitudinal fibrils in the cytoplasm were intensified, particularly under reduced illumination. In sections treated with Schiff's periodic acid reagent the only structures positive for the aldehyde radical were in the basement membranes of the larger blood vessels in the tumor. Examinations of sections prepared with a variety of stains showed a rare cell with transverse striations in the cytoplasm.

The diagnosis was rhabdomyosarcoma of the vagina.

CASE 2 (A.F.I.P. Acc. 490401, Alexandria Hospital No. 183384).—A 37-year-old white married woman had experienced discomfort at the lower part of the vulva for several days before admission to the hospital. One hundred days previously she had been delivered of her second child by low forceps, with episiotomy and repair. At examination eight weeks post partum she was in good health, and the episiotomy wound was well healed. Four weeks later she became aware of discomfort at the vaginal orifice and noted the presence of a mass. On examination the mass was found to be polypoid, measuring about 1 by 4 cm. It was located at the fourchette near the vulvovaginal juncture, slightly to the left of the midline, and 2.5 cm. to the left of the episiotomy scar. It was soft, bright red, and freely movable. The surface was finely granular and did not seem to be covered by mucous membrane. The lesion was excised. One month later complete vulvectomy and bilateral inguinal lymph node dissection were performed. No residual tumor tissue was found at the second procedure. The patient is living, well, and free from evidence of recurrent or metastatic disease two and one-half years after the excision.

Gross Appearance.—The surgical specimen consisted of a polypoid mass and adjacent soft tissue measuring 3 by 2 by 1.2 cm. The surface was rough and granular. It cut with ease, and the cut surface was uniform in texture and pale gray throughout. No orienting landmarks were identified.

Microscopic Description.—The tumor was composed of bipolar cells which varied in size and shape but were more uniform and arranged in better-defined fascicles than those in the previous case. Most of the cells were fairly plump spindle cells with large vesicular nuclei and abundant eosinophilic cytoplasm. Only a moderate number of strap-like forms were seen. Round (or spherical) forms were not infrequent. Occasional syncytial cells were identified, and a few of these were complex and branching. Occasional degenerating cells were seen. The tinctorial properties of the cells comprising the tumor in this case were identical to those seen in Case 1, but the pleomorphism and atypicality were less pronounced. Mitotic activity was somewhat higher than in Case 1, although atypical mitotic figures were fewer. Intercellular substance was not present or detectable by any of the special stains used. Several slides were examined carefully for cells containing cross striations. Only a few such cells were identified. These cells did not differ in any material respect from the remainder of the tumor cells.

The diagnosis was rhabdomyosarcoma of the vulva.

COMMENT

Probably the earliest case of rhabdomyosarcoma of the vulvovaginal region to be described was mentioned casually in 1870 by Rindfleisch,[14] "According as the muscular fibers belong to the smooth or to the striped variety, we follow Zenker in distinguishing between leiomyomata . . . .A large myoma of the retroperitoneal adipose tissue, which I had an opportunity of examining, was made up of

14. Rindfleisch, E.: A Manual of Pathological Histology, translated from the 2d German edition by E. B. Baxter, London, New Sydenham Society, 1872, Vol. 1, p. 177.

transversely striated spindle-shaped cells; so too, a myoma of the vaginal mucous membrane, which showed an obstinate tendency to recur." If there were any further clinical and pathologic documentation, this case might well be acceptable as the first recorded example of such a tumor. As it stands, the report leaves much to the imagination, but it furnishes a clue to the source of the distinction between smooth and striated muscle cell tumors which was made in 1864 by Zenker.

One of the problems that arises in the study of these tumors is the nature of and the appropiate designation for the cell composing the rhabdomyosarcoma. Many writers (Mallory,[15] Stout[11]) use the term rhabdomyoblast, and to some extent usage has sanctioned this word. However, the suffix "-blast" denotes a precursor type of cell, for example, a lymphoblast is the developmental precursor of the lymphocyte; contrariwise, the cells composing rhabdomyosarcomas are not precursors of striated muscle fibers or of anything else. In fact, examination of myotomes in almost any stage of embryogenesis from 12 mm. to 45 mm. shows that the cells from which the skeletal musculature of the body develops are fairly nondescript small bipolar cells which have an ovoid nucleus containing rather evenly distributed chromatin and inconspicuous cytoplasm which has no cross striations. Only later do they tend to form syncytia with the nuclei arranged in tandem formation. If the cells composing rhabdomyosarcoma are not to be construed as precursors, they are evidently cells which have deviated from their position in the ontogenetic genealogy, much as the cells in a lymphosarcoma (which are not lymphoblasts) have become lymphosarcoma cells; they function only as cells of the rabdomyosarcoma or of complex tumors with areas of rhabdomyosarcomatous differentiation. They are rhabdomyosarcoma cells, or, if one would coin an unnecessary and cumbersome word, rhabdomyosarcocytes. However, Mallory, Stout, and the other writers who use the term rhabdomyoblast to describe this cell do not use the suffix "-blast" in the restrictive sense of a precursor cell. Because there is no necessary connection between cell behavior in normal histogenesis and in tumor development, their use of the "-blast" form is purely denotative, indicating the cell type in the normal adult which the tumor cell most closely resembles. Mallory seems to prefer the term rhabdomyoblastoma to rhabdomoyosarcoma to denote the tumor composed of these cells. However, in one sense this usage asks the question, "What is a rhabdomyoblast?" Surely one would be hard put to identify such a cell in normal, i. e., non-neoplastic, tissue.

In addition to the suffix "-blast," the prefix "rhabdo-" is worthy of some attention. The usual observation in most lexicons is that it is derived from the Greek word *rhabdos*, a rod. In Liddell and Scott's "Greek-English Lexicon,"[16] *rhabdos* is defined as a rod, wand, stick, or switch, and the combining form *rhabdo-* is used in Greek words meaning fasces, the insignia of the lictor, flagellation, the fluting of columns, measuring rods, et cetera. There is also a word, *rhabdotos*, which is defined as applying to the stripes, streaks, or striations of animals (Latin, *virgatus*); this same word is used to describe the fluting on columns. The definitions all seem to imply a longitudinal denotation to the root, a sense of arrangement of objects parallel to a long axis, specifically a longitudinal striation.

15. Mallory, F. B.: The Principles of Pathologic Histology, Philadelphia, W. B. Saunders Company, 1914, pp. 271 and 343-347.

16. Liddell, H. G., and Scott, R.: A Greek-English Lexicon, Ed. 8, New York, American Book Company, 1897.

When we seek to apply this root to muscle tissue, it can be readily applied to the gross appearance of skeletal muscle to the naked eye; the fasciculated muscle fibers form muscle bundles which run parallel to the long axis of the muscle mass. There is no implication of cross or transverse striation. It is somewhat difficult to apply this notion to the short cross striations seen microscopically in skeletal muscle. It was Zenker [17] who in 1864, in a digression in his classic monograph on the changes found in the abdominal muscles in typhoid fever, first utilized the terms rhabdomyoma and leiomyoma to distinguish between the two varieties of muscle tumor known as myoma or myosarcoma, the terms proposed by Virchow in 1854. The following year Buhl [18] described a tumor under the title "Wahres recidivirendes Myom (Rhabdomyoma Zenkers)," and the term took hold. By 1876, Hénocque [19] was able to write, "The term striocellular myoma, which Virchow employed as a synonym for rhabdomyoma, has not been accepted." From that time on the term rhabdomyosarcoma has been used to designate this type of tumor. Parenthetically, the prefix "rhabdo-" was no newcomer to the scientific vocabulary in the 1860's.[20]

In any event, Zenker selected the root "rhabdo-" as the Greek equivalent of certain German words ("originally a staff, then also a stripe, as in the fluting of columns, but also used for the stripes on clothing, hence from the last meaning, i.e., in reference to clothing, suitable to be carried over for the striations of muscle fibers").

This translation involves turning the Greek root at right angles if one is to apply "rhabdo-" to cross striations, but it is a usuage which has stood the test of almost 90 years, and the terms based upon this root will continue to be used despite the 90-degree angle which Zenker imposed upon its etymologic genealogy. It is of more than passing interest that while devising this term Zenker recognized not only that neoplasms of muscle might be devided into smooth (leio-) and striated (rhabdo-) groups but also that the rhabdomyomas occured in "pure" (rein) forms and in "mixed" (gemischt) forms. He also made it perfectly clear that rhabdomyomas might be either homologous or heterologous in the same sense as we use these terms today.

Cells which are labeled rhabdomyoblasts are generally described as having one of three common appearances—round cells, strap-like cells, and racquet-shaped or tadpole-shaped cells. These cells can be readily identified by their abundant, intensely eosinophilic cytoplasm and their rather large vesicular nuclei, which usually contain a single, prominent, often amphoteric nucleolous. These

17. Zenker, F. A.: Über die Veränderungen der willkührlichen Muskeln im Typhus abdominalis: Nebst einem Excurs über die pathologische Neubildung quergestreiften Muskelgewebes, Leipzig, F. C. W. Vogel, 1864, pp. 84-86.

18. Buhl, L.: Wahres recidivirendes Myom (Rhabdomyoma Zenkers), Ztschr. Biol. **1**:263-272, 1865.

19. Hénocque, A., in Dictionnaire encyclopédique des sciences médicales, 1876; Series 2, Vol. 11, pp. 239-241.

20. The term rhabdomancy was used in the late 17th Century to describe the pseudoscience involving the use of divining rods. In 1658 Napier devised a calculating apparatus composed of two long strips of bone or ivory, much like our present day slide rule; this device was subdivided into measuring units by small lines which ran transversely, perpendicular to the long axis of the strip. The science of using this calculator was known as rhabdology. Possibly this was the point at which the confusion between longitudinal and transverse striations entered the scientific vocabulary.

cells have a tendency to become binucleate or multinucleate, even syncytial, occasionally forming branched fiber-like cells of considerable size. The cytoplasm of these cells is often smooth and homogeneous, even glassy and highly refractile; however, on occasion the cytoplasm may be granular either in whole or in part, or it may exhibit longitudinal myofibrils or cross striations. In addition to these easily identifiable forms, there are probably many such cells in these tumors which are not fully developed and which have nuclei that are round to oval and have varying amounts of cytoplasm which is relatively unstructured and does not stain so brightly with eosin as the more fully developed forms. Many such cells are seen in sarcoma botryoides and are classified as immature mesenchymal cells, for it is not possible to state with any certainty just when a cell has irreversibly been committed by either its heredity or its environment to becoming a rhabdomyoblast.

Stout,[11] who has described these cells with great care, has pointed out the difficulty of demonstrating cross striations by the usual histotechnical methods. Of his 14 cases of rhabdomyosarcoma, only 9 showed longitudinal myofibrils and/or cross striations. It is implicit from this that cross striations, while they are convincing evidence of the nature of the cells composing this type of tumor, are by no means a necessary condition for the diagnosis. Support for this point of view is found in Borst's description of rhabdomyosarcomas in Aschoff's "Pathologische Anatomie," [21] "These tumors are characterized by their content of longitudinally and transversely (often rich in glycogen) muscle bundles and fibers, largely undifferentiated (spindle cells with transverse striations). Longitudinally striated and nonstriated (smooth) spindly, polymorphous, even round cells are nondifferentiated muscle elements; sometimes also tubular structures occur with peripherally striated differentiation and central undifferentiated nucleated sarcoplasm (embryonal forms). The (often multiple) nuclei of the myoblast are sometimes superficially and sometimes centrally located (within the sarcoplasm). A sarcolemma is mostly lacking or only suggested. The fibers are arranged in bundles or are irregularly dispersed."

Cross striations, then, are by no means the most constant or significant morphologic feature which can be used to identify the rhabdomyoblast. In fact, in most rhabdomyosarcomas which contain cells with clearly visible cross striations, they are found only in a very small percentage of the cells; yet the cells in which they are not found exhibit all the other stigmata of a rhabdomyoid nature.

In the light of this discussion, the two cases reported are classified as cases of rhabdomyosarcoma. Likewise, the case reported by Sadler and Dockerty [10] would seem to belong in the same classification. It is conceded that the term myosarcoma, possibly "rhabdo-," might be preferable for taxonomic purity. The identity of the granular cell "myoblast" with primitive muscle cells is still a matter of controversy, and many observers are not convinced that the granular cell myoblastoma is of muscular origin.

The problem of the histogenesis of these tumors has never been satisfactorily solved. McFarland [2] cites the views of Cohnheim on dysontogenesis and of Wilms on mixed tumors; he is willing to let the matter rest there, deriving the heterologous tumors of the female genital tract from pluripotent mesenchymal elements associated with the development of the wolffian and müllerian ducts. In a recent report

21. Borst, M.: Echte Geschwülste, in Aschoff, L.: Pathologische Anatomie, Ed. 4, Jena, Gustav Fischer, 1929, Vol. 1, p. 814.

on rhabdomyosarcoma of the corpus uteri, Kulka and Douglas [13] cite a variety of theories of histogenesis, among them the ones we have mentioned; these theories include Pfannenstiel's concept of metaplasia from tissues normally present, Robert Meyer's hypothesis that "illegal cell connections" exist between the caudal mesoderm and the urogenital ridge, and Lahm's modification of Wilms's concept of the role of the Wolffian ducts to apply to the Müllerian ducts as well. However, what is applicable to the uterus need not prevail in the vulvovaginal area. Countiss,[22] in his discussion of the paper by Sadler and Dockerty,[10] pointed out that the homologue of the cremasteric muscle is found in the labia majora, and that rhabdomyosarcomas can develop from it. This supports Pfannenstiel's theory of metaplasia from elements normally present. However, Countiss did not exclude the possibility of origin from displaced dysontogenetic cells.

One cannot exclude metaplasia from consideration in rhabdomyosarcomas arising in the vulva or labia because of the presence of the homologue of the cremasteric muscle, but the genesis of predominantly rhabdomyomatous sarcomas that arise in the upper third of the vagina, where skeletal muscle is not normally found, must be explained in terms of one of the theories concerning the nature of "pure" (as opposed to "mixed") heterologous tumors.

The two tumors described were both fungating growths which apparently manifested themselves clinically in a short period of time. Neither gave evidence grossly or microscopically of invading deeply. Stout has pointed out that some of the rhabdomyosarcomas he has studied are apparently indolent tumors and, if situated near the skin surface, can be excised locally without recurrence. However, the microscopic appearance of these tumors does not differ noticeably from that of rhabdomyosarcomas, which disseminate widely and rapidly. Although no follow-up data are available on the patient in Case 1, the gross and microscopic appearance of the tumor might tend to favor its classification as one of the more indolent variety. The patient in Case 2 is still under observation but has been free from recurrence or metastasis for two years. Whether the development of the tumor was in any way related to the patient's antecedent pregnancy or episiotomy cannot be determined; however, the base of the polypoid tumor was 2 to 3 cm. from the episiotomy scar. It is noteworthy that both patients presented themselves for treatment relatively soon after the onset of symptoms.

SUMMARY

Two cases of rhabdomyosarcoma, one of the vulva and the other of the vagina, are described. Previously reported cases of sarcoma in this region are reviewed and evaluated.

The origin and meaning of the terms rhabdomyoblast and rhabdomyosarcoma are discussed. Although the prefix "rhabdo-" is thought to imply the presence of cross striations, these are not the most constant or diagnostic features of this muscle tumor, and the etymology from the Greek word *rhabdos* does not support the concept of transverse striation.

The relation of these tumors to other sarcomas of the female genital tract is discussed. Rhabdomyosarcomas in this region are usually heterologous tumors. They may occur in "pure" form or be present as one component in a "mixed" tumor.

22. Countiss, E. H., in discussion on Sadler and Dockerty.[10]

INHIBITION OF MATURATION OF DUCK ERYTHROCYTES BY SODIUM SELENITE

The Counteraction of This Effect by Cysteine

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GALVESTON, TEXAS

ANEMIA in mammals, as a result of chronic selenium poisoning, has been reported by Franke and Potter,[1] Moxon and Rhian,[2] and Smith, Stohlman, and Lillie.[3] However, little attention has been given to the ability of the hematopoietic system in mammals to respond after discontinuation of use of selenium. While making preliminary observations of the effect of selenium on nucleated erythrocytes in the duck, it was observed that an anemia developed without any morphological evidence of injury to the erythrocytes in the peripheral blood.

In early studies the decrease in the number of reticulocytes in the peripheral blood of the duck following the subcutaneous injection of selenium suggested that selenium was producing its effect on the hematopoietic system through its action on the development of the erythrocytes. In order to investigate this phenomenon more thoroughly the present study was carried out to observe (1) the effect of selenium on the total number of erythrocytes and the percentage of reticulocytes in the peripheral blood of the duck, (2) the changes that occur in the cells of the bone marrow, and (3) the effect of cysteine hydrochloride on the response of the hematopoietic system to selenium.

Evidence presented here suggests that selenium interferes with some enzyme system related to the maturation of the erythrocytes, most likely at the endothelial to the megaloblast stage. Since the inhibitory effect of selenium on the maturation of the erythrocytes may be counteracted by cysteine hydrochloride, it appears that selenium may interfere with an enzyme system dependent on the sulfhydryl group for its activity.

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From the Laboratory of Experimental Pathology and the Departments of Pathology, Biochemistry, and Nutrition, Medical Branch, University of Texas.

1. Franke, K. W., and Potter, Van R.: A New Toxicant Occurring Naturally in Certain Samples of Plant Foodstuffs: Hemoglobin Levels Observed in White Rats Which Were Fed Toxic Wheat, J. Nutrition **8**:615-624, 1934.

2. Moxon, A. L., and Rhian, M.: Selenium Poisoning, Physiol. Rev. **23**:305-337, 1943.

3. Smith, M. I.; Stohlman, E. F., and Lillie, R. D.: The Toxicity and Pathology of Selenium, J. Pharm. & Exper. Therap. **60**:449-471, 1937.

METHODS AND MATERIAL

White Pekin ducks were used at ages varying from two weeks to adults. They were fed Purina Duck Startena and Growena and were given water ad libitum. Blood for cell counts was obtained from the web of the foot. Hayem's fluid was used as the diluent for the erythrocyte counts. A small drop of blood was placed on a cover glass and immediately covered with a drop of a 1% aqueous solution of brilliant crystal blue for the reticulocyte count. The cover glass was placed on a glass slide and the edges of the cover glass were covered with petrolatum. Counts were made of the reticulocytes per 1,000 red blood cells.

Sodium selenite was used in an aqueous solution containing 0.75 mg. of selenium per milliliter. It was injected either intraperitoneally, subcutaneously, or given orally by stomach tube.

Ten ducks, 21 days of age, were given a total of 23 mg. of sodium selenite by stomach tube over a period of 11 days. Bone marrow was aspirated from the tibia, femur, and humerus immediately after the birds were killed. Smears were made and air dried, after which they were stained with Wright's and Giemsa's stains at pH. 6.8. A differential count was made on 500 cells using the classification of Sabin, Doan, and Cunningham.[4]

Cysteine hydrochloride was administered subcutaneously as a neutral aqueous solution, prepared daily, containing 14.915 mg. cysteine hydrochloride and 7.98 mg. sodium bicarbonate per milliliter. A solution of phenylhydrazine hydrochloride, prepared by dissolving 1 gm. in 50 ml. of methyl alcohol, was given by stomach tube. The route, frequency, and quantity of sodium selenite, phenylhydrazine hydrochloride, and cysteine hydrochloride are shown in the different experiments.

EFFECT OF SELENIUM ON ERYTHROCYTES AND RETICULOCYTES IN PERIPHERAL BLOOD

Two adult ducks, numbers 281 and 282, were given 2.5 ml. of selenium subcutaneously daily for two days and on the third day, 3.0 ml. Duck 281 died on the third experimental day. On the fourth day 4.0 ml. of selenium was given to Duck 282, and three hours later this bird was moribund and was killed. The effect of selenium on the number of erythrocytes and on the number of reticulocytes in the peripheral blood of these two ducks is shown in Chart 1. It will be noted that there is a diminution in the percentage of reticulocytes in the peripheral blood of both these ducks. The decrease in reticulocytes is greater in Duck 281 than it is in Duck 282.

The above experiment was repeated on two younger ducks, numbers 285 and 286, aged 20 days, and identical results were found, as shown in Chart 1. In these experiments it was shown that a rapid decrease occurred in the percentage of reticulocytes in the peripheral blood following the subcutaneous injection of large amounts of selenium. The decrease in reticulocytes was accompanied by the progressive development of an anemia.

To observe the effect of smaller doses of selenium over a longer period of time, four ducks were observed for 17 days. Two of the birds, numbers 250 and 255, were given 1.0 ml. of selenium daily for four days. A decrease occurred in the number of erythrocytes and in the percentage of reticulocytes in the peripheral blood (Chart 2). Immediately after the discontinuation of the selenium, the percentage of reticulocytes and the number of erythrocytes progressively increased. The peak for the reticulocytes occurred on the sixth day, and thereafter they decreased progressively in number until the normal level was reached on the 14th experimental

4. Cunningham, R. S.; Sabin, F. R., and Doan, S. A.: The Development of Leukocytes, Lymphocytes, and Monocytes from a Specific Stem Cell in Adult Tissue, Contrib. Embryol. **16**:227-276, 1925.

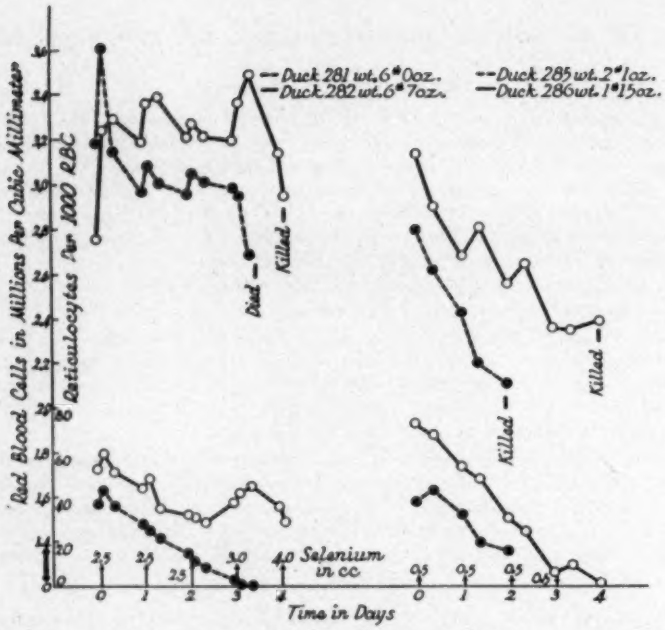

Chart 1.—There occurs a decrease in the number of reticulocytes in the peripheral blood when large doses of sodium selenite are given subcutaneously.

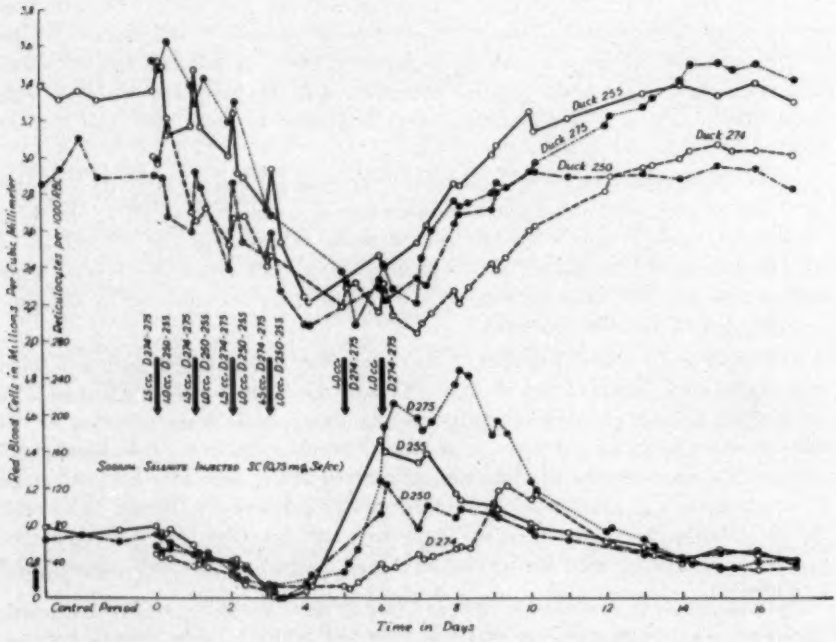

Chart 2.—A decrease occurs in both erythrocytes and reticulocytes in ducks given sodium selenite. This decrease occurs during the first 3 to 4 days, and then there occurs a progressive increase in both reticulocytes and erythrocytes, regardless of the continuous administration of selenium, as shown by Ducks 274 and 275. After this rise the counts return to normal levels.

day. The number of erythrocytes likewise returned to the normal level by the 14th experimental day. Next, two ducks of similar age, numbers 274 and 275, were given 1.5 ml. of selenium daily for four days and 1.0 ml. on the fifth and sixth days, as shown in Chart 2. The percentage of reticulocytes decreased progressively during the first 4 days, and then they began gradually to increase in number. The peak occurred on approximately the 9th day, and thereafter the percentage of reticulocytes decreased progressively until the normal level was reached on the 14th experimental day. The erythrocyte count in these two ducks (numbers 274 and 275) was essentially the same as in ducks 250 and 255 (Chart 2). It is of interest to observe that the percentage of reticulocytes in the peripheral blood started to increase after the

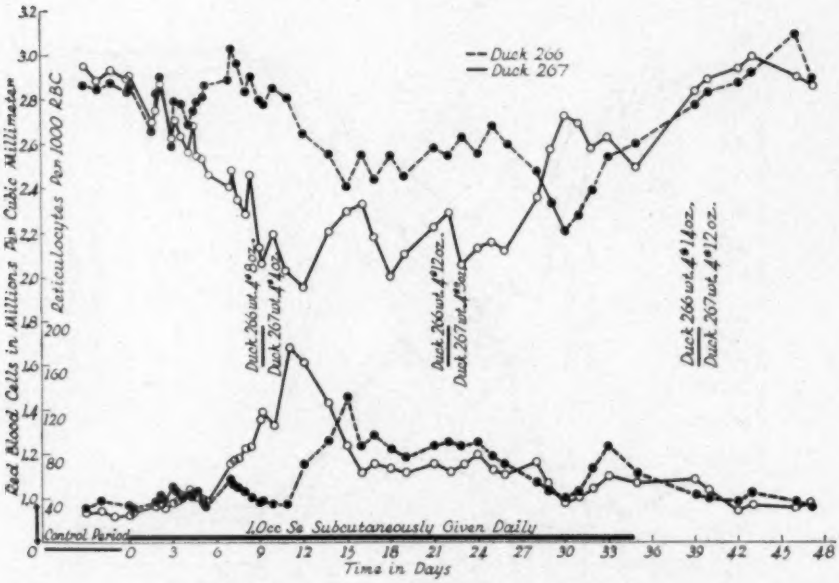

Chart 3.—With small daily subcutaneous injections of selenium, the total erythrocyte count and the number of reticulocytes progressively rise and subsequently gradually decrease. There is a slight anemia in the blood stream as long as the selenium is given.

fourth day while selenium was still being administered. This increase may have been due to the fact that too little selenium was given after the fifth day. It is apparent from the experiments shown in Charts 1 and 2 that the number of reticulocytes and the number of red cells decreased after the subcutaneous injection of large amounts of selenium. Furthermore, the data presented in Chart 2 show that the number of erythrocytes and the number of reticulocytes subsequently return to normal levels when the administration of selenium is discontinued.

Large doses of sodium selenite were administered to the ducks shown in Charts 1 and 2. To the two ducks listed in Chart 3, a smaller dose of selenium (1.0 ml.) was administered daily for 34 days and then discontinued. The number of erythro-

cytes and the number of reticulocytes are shown in Chart 3. A mild anemia developed during the time the selenium was being given; however, when it was discontinued, the red cell count quickly returned to normal level. The number of reticulocytes increased slowly during the first 6 days and reached a peak between the 12th and the 15th day. Thereafter the number of the reticulocytes was slightly elevated as long as the selenium was given; however, when it was discontinued, the reticulocyte count returned rapidly to normal levels.

In the preceding experiments the number of reticulocytes increased in the peripheral blood, although the duck continued to receive selenium. To determine

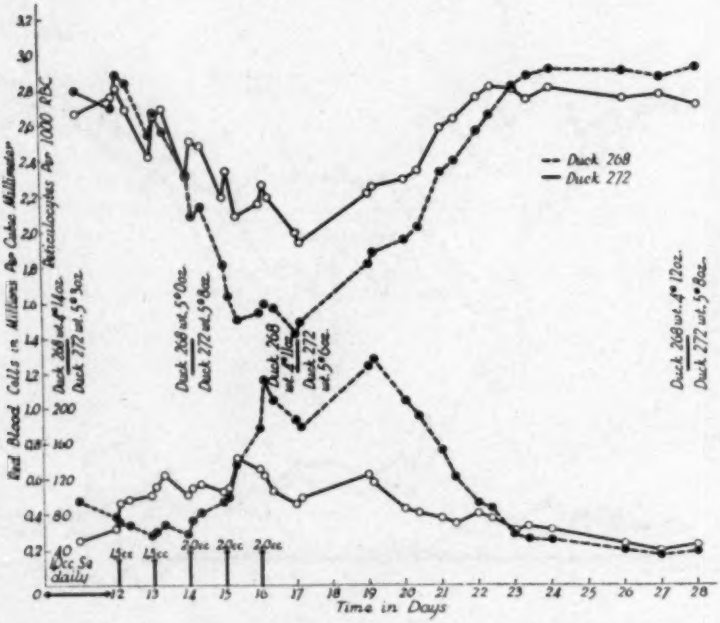

Chart 4.—There occurs a progressive increase in the number of reticulocytes, even when large amounts of sodium selenite are given.

whether this might be explained by the lack of an adequate amount of selenium, two adult ducks, numbers 268 and 272, were given 1.0 ml. of selenium daily for 12 days. Then the dose of selenium was increased to 1.5 ml. for the next two days, and next it was increased to 2.0 ml. for the three succeeding days. At that time all injections of selenium were discontinued. The changes observed in the erythrocyte count and in the number of reticulocytes in the peripheral blood are shown in Chart 4. In these two ducks the number of reticulocytes increased with the increase in the amount of selenium given. The degree of the anemia continued to increase as long as the selenium was given. When the drug was discontinued, both the number of reticulocytes and the number of red cells returned to normal levels.

EFFECT OF PHENYLHYDRAZINE HYDROCHLORIDE AND SODIUM SELENITE ON ERYTHROCYTES AND RETICULOCYTES IN PERIPHERAL BLOOD

From the preceding experiments it may be concluded that sodium selenite inhibits the formation of erythrocytes; however, this inhibitory action is never sufficient to overcome the stimulus to the bone marrow that results from an anemia. To illustrate this phenomenon and also to determine if there is quantitative evidence of an inhibition in the response of the bone marrow of selenium-treated ducks when a hemolytic anemia is produced, the following experiment was performed: Two ducks, numbers

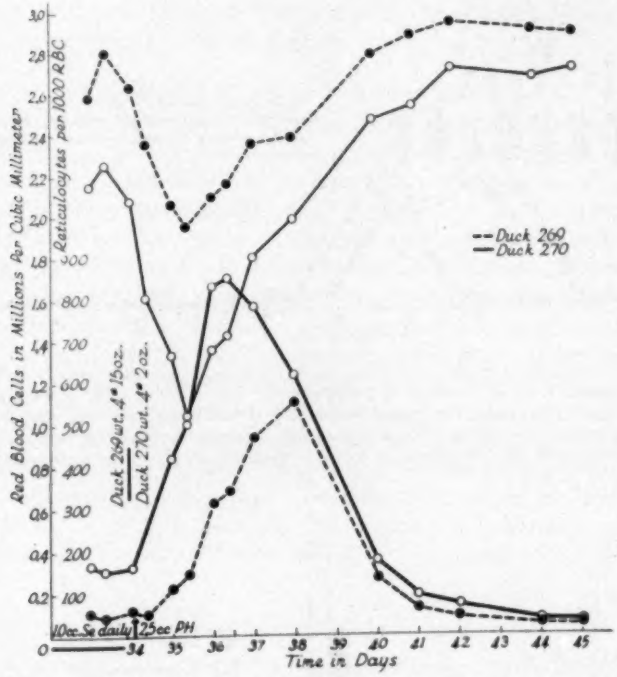

Chart 5.—These two ducks were given sodium selenite daily for 34 days; following this, a hemolytic anemia was produced with phenylhydrazine hydrochloride. The hematopoietic system responded with many reticulocytes.

269 and 270, were given selenium for 34 days, followed by a dose of phenylhydrazine hydrochloride (Chart 5). There occurred a severe anemia accompanied by a larger number of reticulocytes. It is evident that the bone marrow in these ducks receiving a small dose of selenium has the ability to respond adequately to the stimulus resulting from the hemolytic anemia produced by the phenylhydrazine hydrochloride.[5]

To determine if a quantitative difference could be shown in the response of the bone marrow of ducks treated with selenium and given phenlhydrazine, two ducks,

5. Rigdon, R. H.: Acute Anemia Produced by Phenylhydrazine Hydrochloride in the Duck: Observations on Nucleated Erythrocytes in Vivo, to be published.

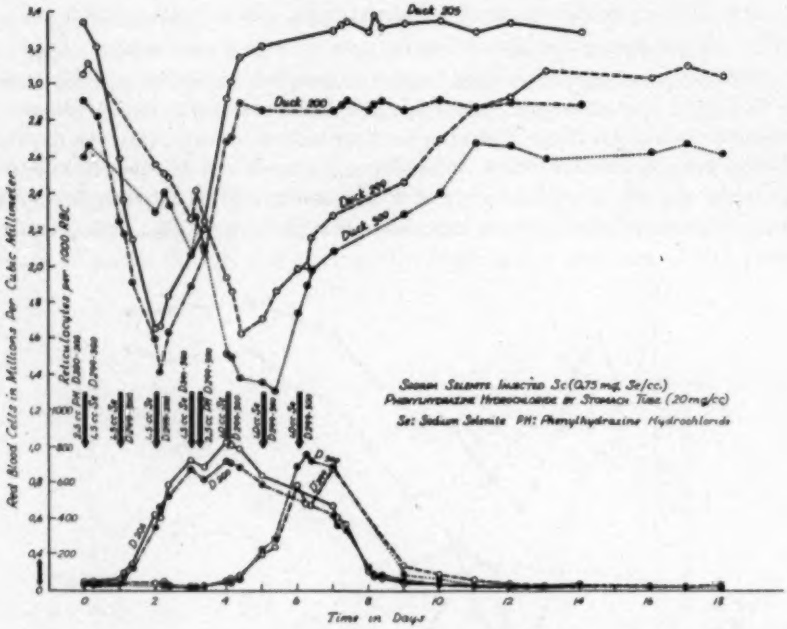

Chart 6.—Ducks 200 and 205 were given phenylhydrazine hydrochloride. Note the development of an anemia and the response in reticulocytes. Ducks 299 and 300 were given selenium for four days and then given the phenylhydrazine hydrochloride. Selenium was also given for the following three days. A hematological response occurred in the latter two birds similar to that in Ducks 200 and 205. The selenium does not affect the ability of the duck to respond to the anemia.

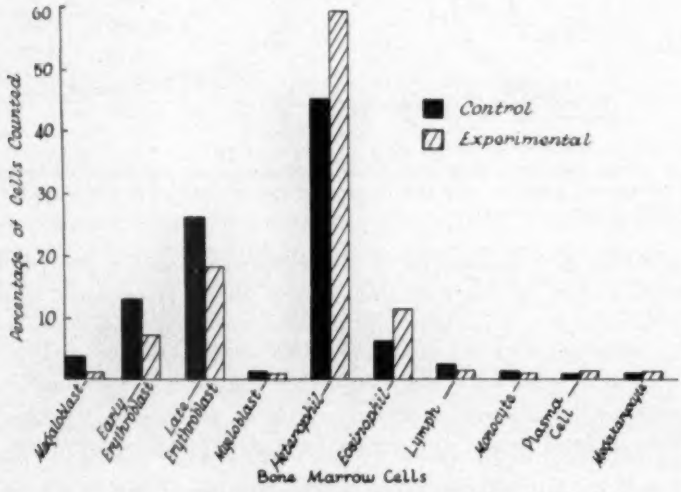

Chart 7.—Marrow of ducks given sodium selenite.

numbers 200 and 205, were given one dose (2.5 ml.) of phenylhydrazine hydrochloride. The typical changes occurred in the number of erythrocytes and reticulocytes, as shown in Chart 6. Two ducks, numbers 299 and 300, were given a medium-size dose of sodium selenite and a smaller dose (1.5 ml.) of phenlhydrazine hydrochloride. One milliliter of sodium selenite also was given to these two ducks for the succeeding three days. The anemia increased rapidly in severity after the administration of phenylhydrazine. The percentage of reticulocytes increased progressively, as shown in Chart 6. The number of erythrocytes and reticulocytes returned to normal levels, however, after the discontinuation of all medication. From these experiments we were unable to demonstrate any inhibitory effect of selenium on the number of reticulocytes that enter the peripheral blood in response to the anemia produced by phenylhydrazine.

BONE MARROW CHANGES PRODUCED BY SELENIUM

Observations on the peripheral blood of ducks given selenium suggest that an anemia occurs as a result of some interference in the maturation of erythrocytes in the bone marrow. This interference phenomenon, as produced by selenium in the formation of erythroblasts, is not absolute but relative. It may be overcome by the development of an anemia. There is no morphological evidence to indicate that the erythrocytes are destroyed within the peripheral blood through the action of selenium. There was no evidence of hemolysis in the ducks given sodium selenite.

The differential count of the cells in the bone marrow of six ducks given sodium selenite is shown in Chart 7. Selenium produces a hypoplasia of the marrow. This hypoplasia is selective for the erythroid series. The decrease in the number of erythroid cells is most marked at the megaloblast level, which suggests an inhibitory effect of the selenium on the maturation of the endothelial cell to the megaloblast. There is a decrease in the number of mitotic figures in the erythroblasts. No abnormal cells were noted. Selenium had no depressant action on the granulocytes or megakaryocytes in the bone marrow. The erythroid-myeloid ratio was 1:1.4 in normal ducks 21 days of age,[6] while it was 1:3 in ducks of similar age given selenium.

EFFECTS OF CYSTEINE ON HEMATOPOIETIC CHANGES PRODUCED BY SODIUM SELENITE

Since McConnell[7] had observed that cysteine hydrochloride inhibits the lethal action of selenium when given to rats, an experiment was planned to determine the effect of cysteine on the hematological changes resulting from the action of selenium on the duck. Two ducks were given 1.0 ml. of cysteine daily for eight days and two doses of 1.0 ml. each for the following two days. No change occurred either in the total erythrocyte count or in the number of reticulocytes per 1,000 red blood cells. Four ducks were then given a subcutaneous injection of cysteine and one of selenium at approximately the same time but in different areas of the body. No significant variations were observed in the cysteine- and selenium-treated ducks when compared with the four controls receiving only the selenium.

6. Crass, G., and Rigdon, R. H.: Bone Marrow in the Normal White Pekin Duck, to be published.

7. McConnell, K. P.: The Effect of Cysteine on the Toxicity of Selenium in Rats, unpublished data.

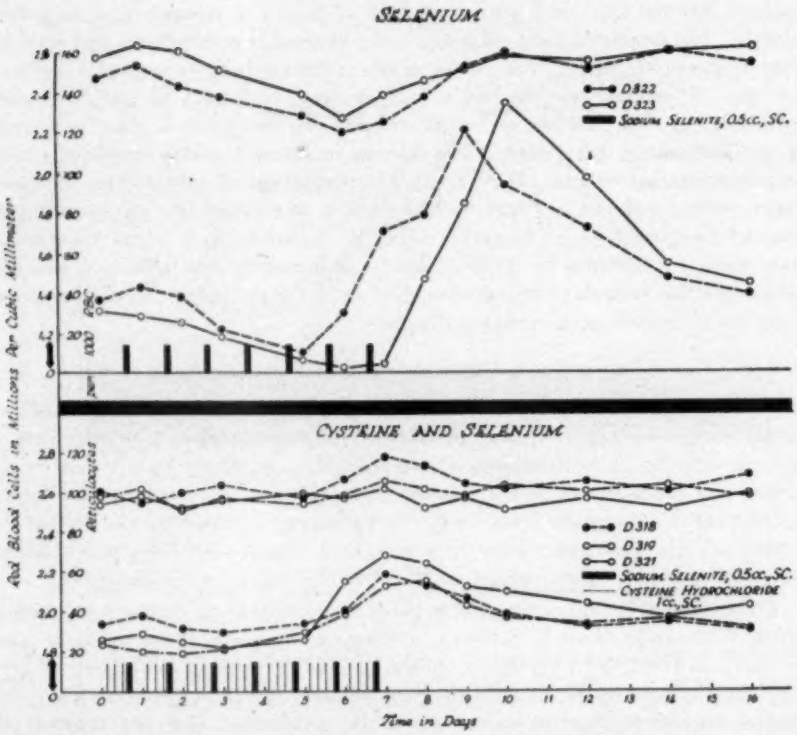

Chart 8.—A decrease in the number of reticulocytes occurs following large doses of selenium, as shown by Ducks 322 and 323. This effect is prevented when large amounts of cysteine are given. In each graph the curve for the total red cell count is placed above the curve, showing the effect on the number of reticulocytes.

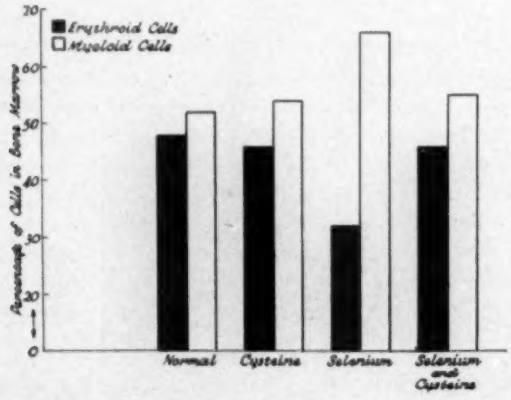

Chart 9.—The percentage of erythroid cells in the bone marrow is approximately the same in the normal ducks as in the birds given cysteine and in those given cysteine and sodium selenite. Note the decrease in the erythroid cells in the ducks given only sodium selenite.

In the next experiment, three ducks, numbers 318, 319 and 321, 26 days of age, were given a larger amount of cysteine in one to three doses preceding the time that the selenium injections were given. Two ducks, numbers 322 and 323, were given only the selenium as shown in Chart 8. There occurred little variation in the total erythrocyte count in the cysteine-selenium-treated ducks during the 16 days of this experiment. A slight diminution did occur in the percentage of reticulocytes during this period. However, the two ducks given only the selenium showed a marked decrease both in the total number of erythrocytes and in the percentage of reticulocytes in the peripheral blood during this period. The experiment was repeated on two ducks given cysteine and selenium and on two ducks given only selenium. The results were similar to those previously described, except for the fact that a slight decrease occurred in the total erythrocyte count in the cysteine-selenium group. The exact dose of cysteine and the optimum time for the administration for the complete inhibition of selenium were not determined. The important factor, however, is to administer the cysteine at least 30 minutes preceding the time of the subcutaneous injection of the selenium. Injections should be made in different anatomic sites to prevent any local reaction between the cysteine and the selenium.

The bone marrow was studied in 12-day-old ducks. Four were given 3.0 ml. of cysteine in three divided doses over a period of six hours each day for seven days. Four ducks were given 0.05 to 0.1 ml. of selenium as a single injection once each day for seven days. Four ducks were given both cysteine and selenium in the same doses as the above for seven days. These 12 ducks were killed on the eighth day of the experiment. The bone marrow from the femur, tibia, and humerus was studied from each of these 12 ducks. A total of 1,000 was counted from the marrow of each of these birds. The results are shown in Chart 9.

It may be seen that the percentage of erythroid cells in the bone marrow is the same in the four ducks given cysteine and the four given cysteine and selenium. Furthermore, the percentage of erythroid cells is the same as that previously observed in normal ducks of similar age.[6] The four ducks given only selenium show a definite decrease in the percentage of erythroid cells when compared with the above two groups of ducks receiving the cysteine and the cysteine and selenium.

COMMENT

In the duck it has been demonstrated that after the administration of sodium selenite the number of reticulocytes in the peripheral blood decreases. Since the selenium effect can be counteracted by cysteine, this suggests that selenium interferes with some sulfhydryl enzyme system related to the maturation of the duck erythrocytes. It is well known that many metals combine with sulfhydryl groups of proteins to form mercaptides.[8] If metals combine with sulfhydryl groups of enzymes requiring the presence of the sulfhydryl groups for activity, inhibition results. Since several enzymes are sulfhydryl enzymes, it is reasonable to assume that the toxicity of specific elements is due primarily to inhibition of the sulfhydryl enzymes. While it is not definitely known, it is possible that the effect of selenium on the hematopoietic system may be due principally to the inhibition of sulfhydryl

8. Barron, E. S. G., and Singer, T. P.: Studies on Biological Oxidations: Sulfhydryl Enzymes in Carbohydrate Metabolism, J. Biol. Chem. **157**:221-240, 1945.

enzymes. The inhibition produced by selenium on sulfhydryl enzymes, as with other elements, may be reversed by the action of thiols.[9] Of interest in this connection is the fact that the lethal action of selenium for the rat has been inhibited through the action of cysteine [7] and glutathione.[10]

Although not entirely related to the present discussion, it should be pointed out that the relationship between hemoglobin and selenium (selenohemoglobin) appears to be similar to that of sulfhemoglobin. The fixation of selenium in the red blood cells has been reported by Dudley [11] and McConnell and Cooper.[12] The chemical reaction between selenium and hemoglobin has been discussed by Dudley,[11] McConnell,[13] and Westfall and Smith.[14]

The absence of morphologic changes in the red cells in the peripheral blood would lend support to the idea that the anemia is secondary to the failure of formation of erythrocytes in the bone marrow. Furthermore, the demonstration of radioactive selenium in the bone marrow of ducks would support the suggestion that a reaction could be occurring within the marrow.[15]

The observation that reticulocytes may decrease in number and then increase in the peripheral blood during the time of administration of sodium selenite presents an interesting problem. Furthermore, the appearance of many reticulocytes in the peripheral blood of ducks given selenium when a hemolytic anemia is produced by phenylhydrazine hydrochloride may indicate that the extra medullary tissues in the duck increase their activity following the action of selenium on the bone marrow. The activity of the extramedullary blood-forming tissues in the anemia associated with malaria has been described previously.[16] It would be of considerable interest to establish as a fact whether or not selenium concentrates in the areas of extramedulllary blood formation the same way as it does in the bone marrow.

The morphologic study of the bone marrow has shown a marked decrease in the number of erythroid cells in ducks given the sodium selenite. The decrease in number of reticulocytes in the peripheral blood is only a reflection of this bone marrow change. It is also of interest to note that no abnormal cells were observed in the bone marrow. This observation would support the thesis that the primary

9. Barron, E. S. G., and Kalnitsky, G.: The Inhibition of Succinoxidase by Heavy Metals and Its Reactivation with Dithiols, Biochem. J. **41**:346-351, 1947.

10. Dubois, K. P.; Rhian, M., and Moxon, A. L.: The Effect of Glutathione on Selenium Toxicity, Proc. South Dakota Acad. Sc. **27**:47-49, 1948.

11. Dudley, H. C.: Toxicology of Selenium: A Study of the Distribution of Selenium in Acute and Chronic Cases of Selenium Poisoning, Am. J. Hyg. **23**:169-180, 1936.

12. McConnell, K. P., and Cooper, B. J.: Distribution of Selenium in Serum Proteins and Red Blood Cells After Subcutaneous Injection of Sodium Selenate Containing Radioselenium, J. Biol. Chem. **183**:459-466, 1950.

13. McConnell, K. P., and Martin, R. G.: Biliary Excretion of Selenium in the Dog After Administration of Sodium Selenate Containing Radioselenium, J. Biol. Chem. **194**:183-190, 1952. McConnell and Cooper.[12]

14. Westfall, B. B., and Smith, M. I.: Distribution of Selenium in Plasma and Liver Proteins and Its Fractionation in Tryptic Liver Digests, Pub. Health Rep. **55**:1575-1583, 1940.

15. McConnell, K. P., and Rigdon, R. H.: Distribution Studies in the Duck After Administration of Sodium Selenate Containing Radioselenium, to be published.

16. Rigdon, R. H., and Rostorfer, H. H.: Observations on the Anemia in Ducks Infected with P. Lophurae, Blood **2**:244-255, 1947.

action of selenium is on the maturation of erythrocytes and, furthermore, that this effect takes place in the formation of these cells and does not take place after they are formed.

SUMMARY AND CONCLUSIONS

White Pekin ducks given sodium selenite show an anemia and a decrease in the percentage of reticulocytes in the peripheral blood.

The bone marrow becomes hypoplastic as a result of the effect of selenium on the erythroid series. The erythroid-myeloid ratio is 1 : 1.4 in normal ducks 21 days of age. This ratio is 1 : 3.0 in ducks of similar age given selenium.

The decrease in erythroid cells is most marked at the megaloblast level, suggesting an inhibitory effect on the maturation of the endothelial cells to the megaloblast.

There is a decrease in the number of mitotic figures in the erythroblasts in comparison to the normal duck, but no abnormal cells are noted.

The mechanism of the action of selenium on the hematopoietic system is discussed. It is suggested that selenium interferes with some enzyme system in the formation of erythroblasts.

The effect of selenium on the hematopoietic system is counteracted by cysteine, suggesting that the selenium inhibits an enzyme system dependent on the sulfhydryl for its activity.

FREE CYSTS OF THE PERITONEAL SPACE

Report of Four Examples Disclosed at Operation or Autopsy

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AND
M. M. HELPERN, M.D.
FRAMINGHAM, MASS.

OUR INTEREST in the condition here presented was first aroused when one of us (Tedeschi) was consulted on an operation concerning the findings of a gelatinous cyst, about 1 cm. in diameter, found free in the abdominal cavity of a 24-year-old woman with chronic pelvic inflammatory disease. Although it was apparent that the finding was incidental and perhaps unimportant and not related to the main condition which led to the operation, the surgeon requested microscopic examination. The wall of the formation was found to consist of regularly aligned, flattened or low cuboidal cells which lay upon a thin layer of fibrous connective tissue. Since doubts persisted on the exact nature of the cyst, the report was filed under the heading of "free cyst of the peritoneal space."

Six months had elapsed when we were faced with an identical finding at an operation, and two additional examples were observed thereafter at autopsies.

REPORT OF CASES

CASE 1.—A 24-year-old white woman with chronic pelvic inflammatory disease had had no previous operations. Her past history was not significant.

CASE 2.—A 36-year-old white woman complained of metrorrhagia. Her uterus and left ovary were removed, revealing atypical endometrial hyperplasia with microcystic ovary. There were no previous operations. Her past history was not significant.

CASE 3.—At autopsy, a 71-year-old white man was found to have active bilateral pulmonary tuberculosis and arteriosclerotic heart disease. His past history was not significant, except for the tuberculous process which was recognized about one year prior to death. There were no previous operations.

CASE 4.—At autopsy, a 70-year-old white woman was found to have coronary artery insufficiency, extensive myocardial scarring, and healed apical infarction. Her past history showed anginal attacks and progressive heart failure of two years' duration. She had had no previous significant diseases or operations.

MORPHOLOGY OF THE CYSTS

The appearances of the cysts, both on observation with the naked eye and in the microscopic sections, were almost identical. They ranged in size from 1.2 to 2 cm. in the largest dimension and in shape from round to oval. The contents of

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the cysts were jelly-like and yellowish gray in color. The cystic walls were thin, transparent, smooth, and glistening, both inside and out. In all instances the cyst walls were lined by a single layer of regular, flattened, spindle-shaped, or low cuboidal cells (Fig. 1). In some areas a few cells were missing, and occasionally a larger segment of cyst wall was devoid of cellular layer. In places, the connective tissue fibers of the cyst walls were in compact arrangement, but in other places,

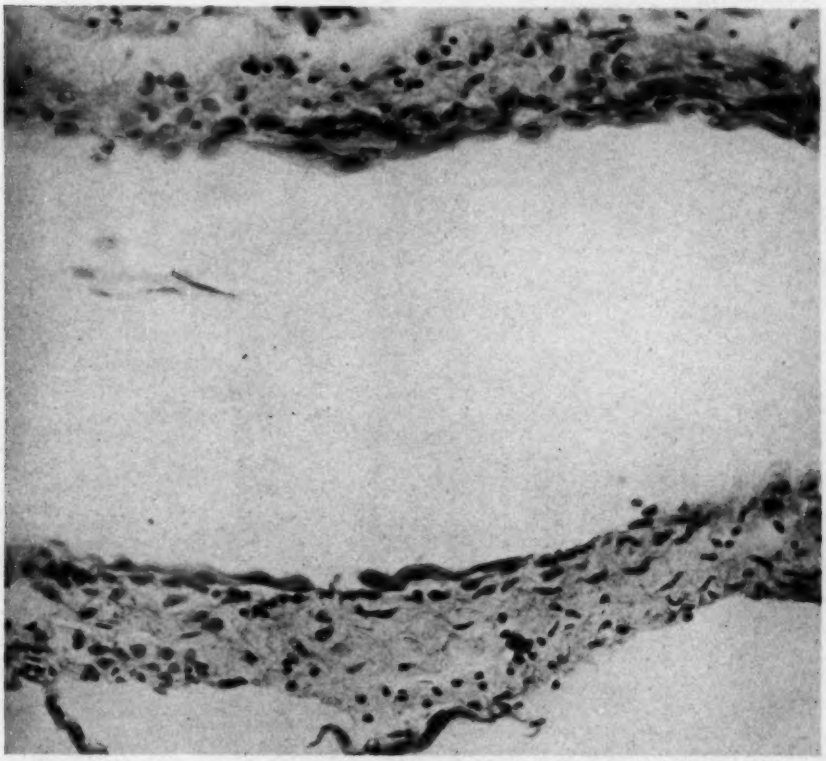

Fig. 1.—One of the cysts found free in the peritoneal space (Case 1) showing a regular lining of flattened or low cuboidal cells lying upon a thin layer of collagenous tissue. Nearly identical structure was displayed by the other "free" cysts included in this report; × 200.

COMMENT

they were spread apart apparently because of edema. No cellular elements were present in the lumen of the cysts.

An attempt to ascertain the nature of these cysts found free in the peritoneal space leads to the same difficulties that are encountered in interpreting the nature of other intra-abdominal cysts. In the cases under consideration, the difficulty is increased by the fact that the cysts were not connected with the organ or structure from which they may have originated.

From the time of Benevieni's [1] description of a cyst in the mesentery, casually discovered at autopsy and considered to be an anatomical curiosity (1507), many attempts have been made to classify in an orderly fashion the comparable conditions gathered thereafter in the medical literature.

The generally accepted subdivision into congenital and acquired cysts [2] is here introduced as a starting point in the discussion of the nature and of the most likely site of origin of the cysts found free in the abdominal cavity in our four patients.

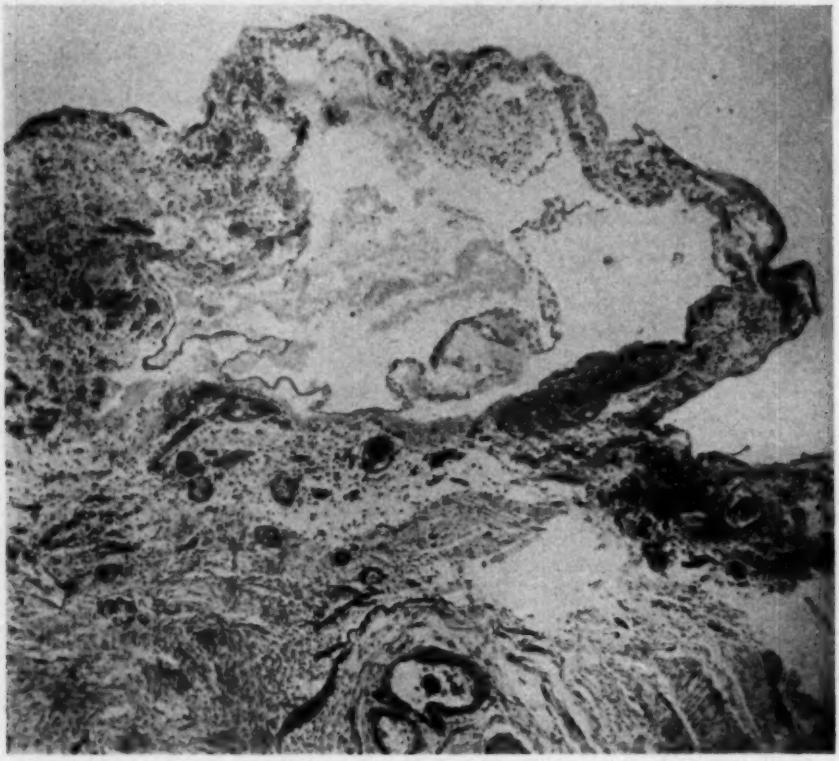

Fig. 2.—Subserous chylous cyst of the appendix; × 80.

The enteric cysts make up a large group among the cysts of congenital origin.[3] They are most frequently found in proximity to the ileocecal valve, and their intestinal origin is revealed by the presence of the usual components of the intestinal wall, including the mucosal layer and the muscular coat. In view of the basic differences in structure, it is apparent that none of the cysts under present consideration can fit in this category.

1. Benevieni, cited by Beahrs, O. H., and Judd, E. S., Jr.: Chylangiomas of the Abdomen, Proc. Staff Meet., Mayo Clin. **22**:297, 1947.

2. Warren, K. W.: Peritoneal Cyst, Lahey Clin. Bull. **5**:185, 1947.

3. Grimes, A. E.: Mesenteric Cysts, Am. J. Surg. **77**:528, 1949.

On the basis of structural differences, it can be stated that the present cysts had not originated from aberrant suprarenal tissue since cysts of this sort are characterized by the presence of cortical suprarenal cells. On similar ground, it is apparent that the cysts we have examined are not related to the inclusional dermoid cysts, the cystic teratomas, the cysts derived from germinative epithelium, the duct of Müller, or the coelomic epithelium.

Cysts of Wolffian origin also belong to the category of the congenital cysts. They are characterized by a regular lining of tall columnar or low cuboidal cells lying

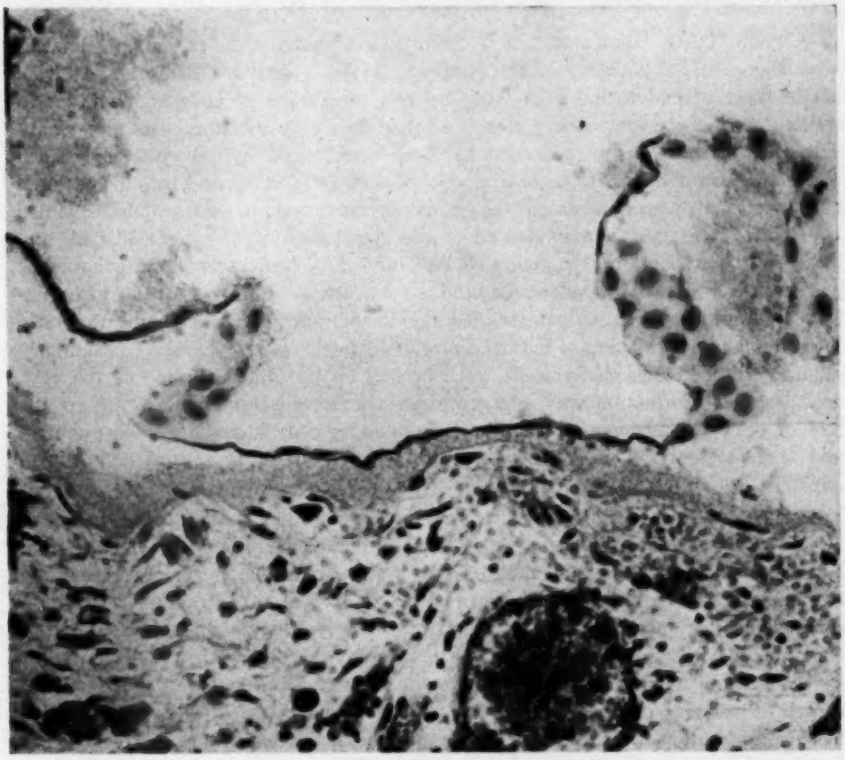

Fig. 3.—Subserous chylous cyst of the appendix (Fig. 2) showing at higher magnification a structure very similar to that disclosed by the cysts found free in the peritoneal space; × 200.

upon a thin layer of collagenous tissue. Since flattening of the epithelial lining can occur as a result of intraluminal pressure, some thought must be given to the possibility that the cysts under discussion might have been Wolffian cysts detached from their site of origin.

As great a variety, or perhaps even a greater variety, of acquired cysts has been recognized. Among these, the best known are the pneumatic, hematic, and parasitic cysts and the cysts derived from lymph channels.

Absence of air, blood, or parasites indicates that the cysts under consideration do not belong in any of these categories.

Chylangiomas arising in the mesentery, intestine, omentum, and retroperitoneal space have been described,[4] and they were generally interpreted as hamartomas of the system of the lymph channels. From our own files, three examples of chylangiomas, one from the mesentery and two from the retroperitoneal space, were reviewed and compared with the free intraperitoneal cysts. In the former group, the tumor masses were multilocular and consisted of cavernous lymph spaces lined by flattened, unstratified, or hyperplastic budding endothelium heaped up into layers. Elastic tissue and a few bundles of smooth muscle were present in the walls of the cysts, the contents of which were chylous. These obviously were not the characteristics of our free cysts.

Chylous cysts, from obstruction or imperfect canalization of lymph channels, also belong to the category of the lymphatic cysts. They are characterized by a single layer of endothelial cells lying upon a membrane of collagenous tissue, a structure which compares very closely to that shown by the free cysts under consideration. Although no indication has been found that chylous cysts can detach themselves from their site of origin, this possibility is suggested by our personal observation of a pedunculated subserous chylous cyst in an acutely inflamed appendix. This cyst was implanted at the apical region, was dome-shaped, and measured about 1 cm. in diameter. Its wall was thin, transparent, and gelatinous; golden-yellow material was contained in the cyst's cavity. On microscopic examination, this cyst was seen to consist of a thin membrane of fibrous tissue regularly lined by one or two layers of flattened or cuboidal cells (Figs. 2 and 3). Similarly aligned flattened mesothelial cells were present in the outer surface of the cyst, and the whole formation was interpreted as a subserous lymphatic cyst. At the base of the cyst there were clusters of inflammatory cells, abundant blood pigment, and areas of scarring, in the proximity of which there were several endometrial implants.

Additional sections showed that the peduncle of the cyst consisted of a dilated lymph channel, partially lined by flattened or cuboidal epithelium, which ran in a tortuous fashion in the appendiceal serosa and outer muscular coat. At that time it was clear that the chylous cyst had resulted from partial obstruction of a lymph channel, brought about by the inflammatory reaction elicited by the endometrial implants. It was furthermore possible to rule out the theory that the cyst might have been of peritoneal origin, according to a mechanism outlined by Hertiley,[5] namely, formation of fibrinous exudate over a granular mass, followed by endothelial proliferation, which results in the formation of a new peritoneal leaflet. That this appendiceal cyst might have become a "free" cyst by detachment from its lymphatic stem was also a foreseeable occurrence. In view of the structural similarities, it is not unlikely that the free cysts in our study had a similar pathogenetic mechanism.

Since the cystic lesions reported here were devoid of blood supply, one might have expected to find evidence of regressive changes. Instead, the cellular structures in all four cysts were well preserved, and this indicates that they must have been nourished through exchange of substances in peritoneal fluid.

4. Lubitz, J. M., and Flynn, R. W.: Chylangioma Cavernosum Mesenterii, Surgery **18**:772, 1945.

5. Hertiley, A. E.: The Peritoneum, St. Louis, C. V. Mosby Company, 1919.

SUMMARY AND CONCLUSIONS

The occurrence of free cysts in the peritoneal space is reported. In two instances the lesion was observed during operations, and in the two remaining instances the cysts were found at autopsies. Three of the patients were women, and one was a man. In all instances the cysts were regarded as incidental findings.

The gross and microscopic appearances were nearly identical in all four examples. The walls of the cysts were comprised of a thin layer of collagenous fibers supporting a lining layer of flattened, spindle-shaped, or low cuboidal cells.

A comparison between the structure of these "free" cysts and the structure of other attached intra-abdominal cysts leads us to suggest that the former may originate by detachment from either Wolffian cysts or chylous cysts.

Case Reports

COEXISTING CONGENITAL STENOSES OF AORTIC AND PULMONIC OSTIA

GOETZ W. RICHTER, M.D.
NEW YORK

CONGENITAL stenoses of either the pulmonic or the aortic ostium are relatively common, but their coexistence in one person has apparently never been recorded before. The present report deals with the findings in a patient with congenital heart disease who lived to adulthood and who, after death from other causes, was found to have stenoses of the pulmonic and aortic orifices, together with a patent foramen ovale, a patent ductus arteriosus, a hypoplastic aorta, and syndactylism.

REPORT OF A CASE

A 22-year-old Latvian was admitted to the New York Hospital with chief complaints of heart trouble and blindness. The family history was unremarkable. His mother reported that he was born "a blue baby" and that he had had heart murmurs since birth. He was always incapable of taking exercise because of resultant dyspnea and palpitation. Four years prior to admission reading difficulties developed, and he was found to have bilateral optic atrophy, which was thought to to be due to a suprasellar tumor. An operation was refused. On admission he was in no distress and was not cyanotic. The forehead was unusually prominent. There was webbing of several toes. He had a thoracolumbar kyphoscoliosis. The temperature was normal, the pulse 86 beats per minute. The respirations numbered 18 per minute. The blood pressure in the right arm was 118/72 and in the right leg 160/98. The heart was not enlarged to percussion or to palpation. A systolic thrill was felt in the pulmonic area, and a systolic murmur was audible over the entire precordium. This murmur was loudest in the pulmonic area and was transmitted to the neck, to the left axilla, and to the back. No diastolic murmur was heard. The left testis was undescended. Ophthalmological examination revealed bilateral secondary optic atrophy and papilledema in the right eye. The neurological examination was noncontributory.

Electrocardiograms showed patterns indicative of right ventricular hypertrophy with sinus tachycardia. Angiocardiographic studies indicated the presence of a stenosis in the region of the pulmonic valve but failed to reveal septal defects. X-rays of the skull showed a deepened pituitary fossa. The erythrocyte count was 5,700,000 per cubic millimeter; the leucocyte count was 8,900 per cubic millimeter, with a normal differential count. The urine was normal. On lumbar puncture the spinal fluid pressure was found to be 550 mm. of water. The spinal fluid contained 149 mg. of protein per 100 cc., and had a normal blood cell count.

A craniotomy was performed, and a tumor was found about the tuberculum sellae turcicae. The tumor extended into the pituitary fossa and into the two frontal fossae. It measured approximately 6 cm. in diameter. It was resected and on further examination proved to be a psammomatous meningioma. After the operation the patient failed to regain consciousness and died on the 17th postoperative day. A complete autopsy was performed, but only the pertinent findings will be considered here.

The body was somewhat underdeveloped for the patient's stated age of 22 years. It weighed 44 kg. and measured 159 cm. in length. The heart was normally situated but somewhat dextrorotated. It weighed 350 gm. The epicardial surface was unremarkable. The right auricle was moderately dilated, and its wall was thickened. The foramen ovale was widely patent,

From the Department of Pathology, New York Hospital-Cornell Medical Center.

admitting a probe with a diameter of 1 cm. The wall of the right ventricle was markedly hypertrophied, measuring 10 to 12 mm. in thickness (normal, 2 to 5 mm.). The tricuspid valve measured 8 cm. in circumference and was unremarkable. The pulmonic ostium measured 0.5 cm. in diameter and 2.0 cm. in circumference, having about one-fourth the normal width (normal circumference, 8 to 9 cm.). The cusps of the pulmonic valve were all present but were somewhat smaller than normal. They were slightly thickened and irregular, but their commissures were not remarkable (Fig. 1). Their endocardial surfaces were smooth in spite of the irregularities in the shapes of the cusps. Beyond the pulmonic valve the pulmonary artery became wider, its circumference increasing to a maximum of 6 cm. (normal, about 8 cm.). The pulmonary veins entered the left auricle in the usual way. The auricle itself was normal. The wall of the left ventricle measured up to 18 mm. in thickness. The mitral valve measured 9 cm. in circumference and was not remarkable. The aortic valve ring measured

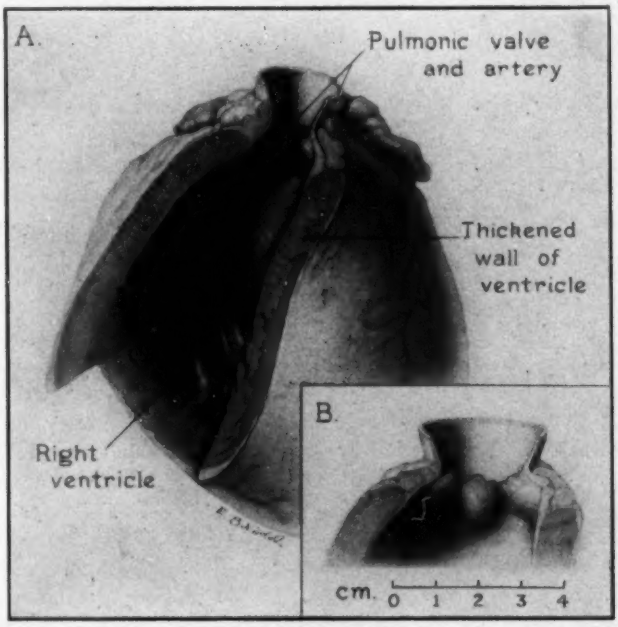

Fig. 1.—Right ventricle and stenotic pulmonic ostium (*A*) and detail of the latter (*B*).

0.8 cm. in diameter and 3.0 cm. in circumference, the ostium thus being less than half as large as normal (normal circumference, 7 to 8 cm.). The cusps of this valve were slightly smaller than normal, considerably thickened, and slightly fused at their commissures (Fig. 2). The sinuses of Valsalva appeared as deep pockets behind the valve cusps. A patent ductus arteriosus with a diameter of 0.8 cm. arose from the aorta just beyond the origin of the left subclavian artery and communicated with the pulmonary artery at its bifurcation. The interventricular septum was intact. The ostia of the coronary arteries were normal, and these vessels had a normal distribution. They showed minimal atherosclerosis.

The entire aorta was underdeveloped, measuring only 4.0 cm. in its widest circumference, in the arch, as compared with a normal average circumference of 7.4 cm. The aorta also showed minimal atherosclerosis.

Examination of the brain disclosed that about one-half of the right frontal lobe had been excised at operation. There was extensive hemorrhage and necrosis in the remaining half of this lobe. This was presumed to have resulted from operative trauma. In the left frontal

lobe there was also extensive hemorrhagic softening, particularly in the region of the second convolution. This could have resulted from inadvertent injury to the left anterior cerebral artery while the tumor was being removed. Further examination of the brain revealed moderate microgyria in both parietal and occipital lobes. There was a mass just anterior to the sella turcica which represented the unexcised portion of the meningioma and which measured 2 by 1 by 1.5 cm.

The lungs weighed 870 gm. together. In the two upper lobes there were scattered gray and red firm areas of consolidation, measuring up to 5 mm. in cross section. Syndactylism of three of the toes in the left foot and an undescended left testis, which was very fibrotic, were also noted.

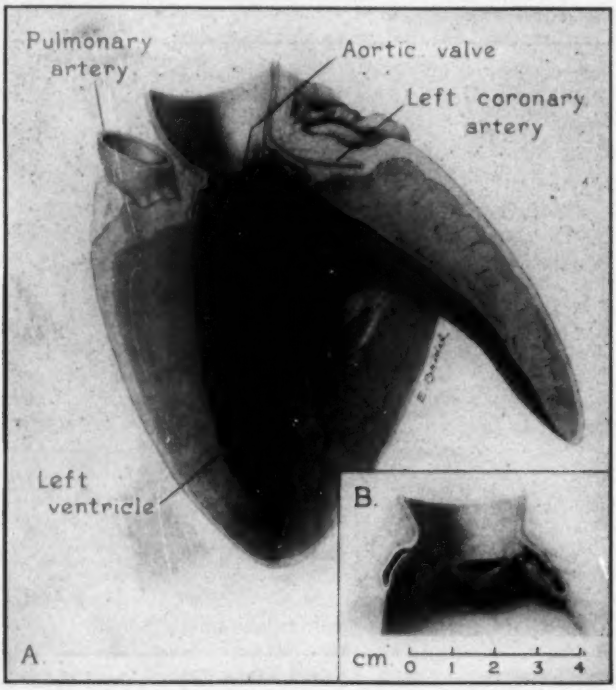

Fig. 2.—Left ventricle and stenotic aortic ostium (*A*) and detail of the latter (*B*).

Microscopic examination revealed marked hypertrophy of the muscle fibers of the right ventricle, together with moderate interstitial fibrosis. There was also some hypertrophy of the myocardium in the right auricle and in the left ventricle. A number of small myocardial arteries and arterioles showed medial hypertrophy and in a few instances intimal hyperplasia. Sections from the pulmonic valve showed a moderate increase in the amount of fibrous and hyaline connective tissue within the valve cusps. Here and there the loose connective tissue had undergone mucinous degeneration. There was a thin layer of elastic tissue underneath the endocardium of the closing surfaces of the cusps. In the cusps of the aortic valve there was a considerable increase of the hyaline and cellular connective tissue, again with scattered mucinous degeneration. Neither of the two valves showed any inflammatory

reaction, and vascularization was altogether absent. The root of the pulmonary artery, behind the valve cusps, proved to be entirely normal microscopically. Beyond the valve, however, the media of the pulmonary artery was thinner than normal, and here it contained somewhat less muscular and elastic tissue than did several normal pulmonary arteries examined for comparison. Sections from the root of the aorta, taken in the region of the sinuses of Valsalva, failed to reveal anything abnormal, and elsewhere the aorta was also unremarkable on microscopic examination.

Microscopic studies confirmed the diagnosis of psammomatous meningioma and that of hemorrhagic softening in the two frontal lobes. In the lungs there was an acute bronchopneumonia.

COMMENT

This seems to be the first reported case of its kind. It is surprising that coexisting congenital stenoses of pulmonic and aortic ostia have not been recorded before, for the segments of the pulmonic and aortic outflow tracts that were involved in the present case develop concurrently in the human embryo, and they do so in close proximity.

The intrapericardial portions of the pulmonary artery and aorta originate from the division of the common truncus arteriosus during the sixth week of fetal life. Four cushions develop in the truncus, and two of these then form the spiral septum that separates the aortic from the pulmonic passage. Keith [1] has stated that a stenosis at the root of the pumonary artery (the infundibulum) may result from an arrest in the development of the pulmonic part of the truncus, whereas certain types of stenosis at the root of the aorta, particularly the so-called subaortic stenosis, may result from a lack of regression of an embryonic fibrous band that is generally located just below the aortic valve cusps. In the present case, however, the basic abnormality is the narrowness of the two circular passages through which blood leaves the heart. It may be considered a hypoplasia of these two passages, particularly since the entire aorta proved to be underdeveloped. Unfortunately, neither the embryologic nor the etiologic basis of these malformations is known. It is impossible to exclude fetal carditis as an etiologic factor, although no scars or other tell-tale remnants of such a process were evident at postmortem examination. On the other hand, the absence of such remnants is evidence against the possibility of a fetal carditis, as Mönckeberg has pointed out in his detailed review of this subject.[2] Moreover, the presence of syndactylism makes it more likely that the cardiovascular abnormalities were the result of a primary developmental defect. Besides, it seems unlikely that hypoplasia of the aorta could result from a fetal carditis.

It should be emphasized that the tightness of the aortic orifice was due entirely to the constriction of the so-called valve ring. The moderate deformities of the aortic valve cusps were not severe enough to constitute a valvular stenosis proper. Though their gross and microscopic appearance was suggestive of an old rheumatic valvulitis, these deformities were an accessory finding. It may be recalled here that congenital malformations in the regions of the heart valves predispose to the later development of valvular deformities in the wake of rheumatic fever.

1. Keith, A.: Fate of the Bulbus Cordis in the Human Heart, Lancet **2**:1267-1273, 1924.

2. Mönckeberg, J. G., in Hencke, F., and Lubarsch, D., Editors: Handbuch der speziellen pathologischen: Anatomie und Histologie, Vol. 2, Berlin, Springer-Verlag, 1924, pp. 1-183.

From an anatomical standpoint the two ostial stenoses were of intermediate severity, and this is probably the reason why the patient survived to adult life. Unfortunately, no cardiac catheterization was performed, and it is therefore not known for certain in which direction blood flowed through the patent foramen ovale and the patent ductus arteriosus. It seems likely, however, that blood passed from the right to the left auricle, because the hypertrophy and dilatation of the former indicated that it had been under greatly increased pressure, doubtless as a result of the pulmonic stenosis. As for the ductus arteriosus, it may be assumed that because of the pressure differential between the aorta and pulmonary artery blood was shunted from the former to the latter. It is conceivable, however, that the ductus arteriosus, though patent, did not carry much blood, for no murmurs indicative of its functioning were heard. Finally, in considering the circulatory difficulties of this patient, it is of interest to recall that coexisting pulmonic and aortic stenoses of an acquired (for example, rheumatic) type may be compatible with life for a long time, as, for example, in a case reported by Dagnini.[3]

SUMMARY

A 22-year-old man with a lifelong history of heart disease was found at postmortem examination to have stenoses of the pulmonic and aortic ostia, together with hypoplasia of the entire aorta, a patent foramen ovale, a patent ductus arteriosus, marked right-sided cardiac hypertrophy, syndactylism of several toes, and an undescended testicle. The two stenoses were due entirely to the narrowness of the roots of pulmonary artery and aorta. This appears to be the first case of coexisting congenital pulmonic and aortic stenoses on record.

3. Dagnini, G.: Doppia stenosi delle valvole polmonari ed aortiche: Ectasia del tronco dell'arteria polmonare a valle della stenosi, Cuore e circolaz. **14**:15-39, 1930.

General Reviews

PATHOLOGY OF EPIDEMIC TYPHUS

Report of Fatal Cases Studied by United States of America Typhus Commission in Cairo, Egypt, During 1943-1945

Prepared by the

COMMITTEE ON PATHOLOGY

DIVISION OF MEDICAL SCIENCES, NATIONAL RESEARCH COUNCIL

With Collaboration of the

ARMED FORCES INSTITUTE OF PATHOLOGY

FOREWORD

HOWARD T. KARSNER, M.D., L.L.D.

THE MANY problems of typhus have preoccupied the minds of those engaged in civilian and military medicine for centuries. This interest was naturally augmented and accelerated with the advent of World War II and especially with the entry of the United States as an active participant in the conflict. Within a few months of the time we were plunged into the war, the Preventive Medicine Service of the United States Army became deeply concerned as to whether, in view of a projected invasion, the control of typhus in North Africa would be adequate, particularly because the modified Cox vaccine was still untried and the field delousing methods, also new, had not yet been applied to troops. Brigadier General (then Colonel) James S. Simmons, Medical Corps, United States Army, took up the matter with his associates in the Preventive Medicine Service of the Army, and the decision was made to include representatives of the Navy and the Public Health Service in further discussions. Consequently a meeting was held Aug. 22, 1942, including three officers from the office of the Surgeon General of the Army, Colonel (later Brigadier General) James S. Simmons, Medical Corps, United States Army,

The Committee on Pathology, National Research Council: Howard T. Karsner, M.D., LL.D., Chairman; Paul E. Boyle, D.M.D.; George R. Callender, M.D.; Russell L. Holman, M.D.; Balduin Lucké, M.D., Dr. P.H.; Sidney C. Madden, M.D. Robert A. Moore, M.D., Ph.D.; Alan R. Moritz, M.D., LL.D.; Benjamin Rones, M.D.; Hilton A. Smith, Ph.D.; Henry A. Swanson, D.D.S.; Shields Warren, M.D., and William B. Wartman, M.D.

Contributions were made by the following: Lieutenant Commander Boyd K. Black (MC), U.S.N., The United States Medical School, Bethesda, Md.; Zola K. Cooper, M.D., Assistant Professor of Pathology, Washington University, St. Louis; Howard T. Karsner, M.D., LL.D., Medical Research Adviser, Bureau of Medicine and Surgery, United States Navy, Washington, D. C.; Ralph D. Lillie, M.D., Medical Director, United States Public Health Service, National Institutes of Health, Bethesda, Md.; William B. McAllister Jr., M.D., Assistant Professor of Pathology, Yale University, New Haven, Conn.; Robert A. Moore, M.D., Ph.D., Dean and Edward Mallinckrodt Professor of Pathology, Washington University School of Medicine, St. Louis; Alan R. Moritz, M.D., LL.D., Professor of Pathology, Western Reserve University, Cleveland; Alwin M. Pappenheimer, M.D., Professor of Pathology, Emeritus, Columbia University, New York; Edward B. Smith, M.D., Professor of Pathology, Indiana University, Indianapolis; David E. Smith, M.D., Assistant Professor of Pathology, Washington University, St. Louis; William B. Wartman, M.D., Morrison Professor of Pathology, Northwestern University, Chicago; Andrew Yeomans, M.D., Chief of the Medical Service, Veterans Administration Center, White River Junction, Vt.

Chief of the Preventive Medicine Service; his Assistant Chief, Lieutenant Colonel S. Bayne-Jones (later Brigadier General), Medical Corps, Army of the United States, and Lieutenant Colonel (later Colonel) William S. Stone, Medical Corps, United States Army, Chief of the Division of Sanitation, together with Captain (later Rear Admiral) C. S. Stephenson (MC), U. S. N., Chief, Division of Preventive Medicine, Bureau of Medicine and Surgery, Navy Department, and Dr. R. E. Dyer, Assistant Surgeon General, United States Public Health Service, Director of the National Institute of Health. Agreement was readily reached to recommend the establishment of a commission to study the various aspects of the problem in the broadest possible manner, to designate it as the United States of America Typhus Commission, and to place it on a high administrative plane. Coordinations were effected largely by Major General (later Lieutenant General) LeRoy Lutes, United States Army, Office of the Assistant Chief of Staff, Army Service Forces, and Colonel William L. Wilson, Medical Corps, United States Army, Chief of Hospitalization and Evacuation Branch, Plans Division, Army Service Forces. Major General A. C. Biggam, Medical Consultant to the British Army, then in Washington, D. C., on a mission, coordinated with the British authorities.

The plan contemplated a joint service organization under the cognizance of the Army. The Secretary of War approved Oct. 22, 1942, and on recommendation of the Surgeons General of Army, Navy, and Public Health Service appointed Captain Charles S. Stephenson (MC), U. S. N., as Director; on similar recommendation he was promoted to the rank of Rear Admiral in November, 1942. Further progress in organization and planning was made at a meeting under the auspices of the National Research Council, with Dr. Wilbur A. Sawyer, Director of the International Health Division, Rockefeller Foundation, in the chair. Military and political channels were followed and on Dec. 24, 1942, the United States of America Typhus Commission was created by Executive Order 9285, signed by the President, Franklin D. Roosevelt.

The Commission was divided into a Forward Echelon and a Rear Echelon, the latter remaining in the United States. The two groups of the Forward Echelon arrived in Cairo, Egypt, Jan. 7 and Jan. 20, 1943, respectively. Here cooperation and aid were rendered by the Ministry of Health of the Egyptian Government, which provided facilities in the Fever Hospitals at Abbassia and Imbaba. Colonel (later Brigadier General) Crawford F. Sams, Medical Corps, United States Army, Surgeon to the United States Army Forces in the Middle East, and Dr. M. A. B. Demerdesh Bey, Director of the Cairo Fever Hospitals, were most helpful. The illness of Rear Admiral Stephenson led to his relief as Director, and on Feb. 8, 1943, Colonel (later Brigadier General) Leon A. Fox, Medical Corps, United States Army, was appointed Director. As time went on, the original membership was supplemented by many other distinguished scientists, but their names cannot be included here. A narrative and detailed history of the Commission is in preparation by Dr. Stanhope Bayne-Jones.

Numerous investigations were carried out under the direction of the Commission in the field in Africa, Europe, Asia, and in laboratories in the United States, including studies of the different forms of typhus and of certain other rickettsial diseases. The practical results of the control measures instituted by the Commission and associated agencies were witnessed especially in the epidemic in Naples

(1943-1944); in the inner Reich of Germany, with its typhus cases spread by and from the concentration camps (1944), and in Japan (1945-1946), with its social and political disorganization and influx of cases from Korea.

As a part of this great program, a study of the pathology of fatal cases was envisioned. Even before Executive Order 9285 was promulgated, Rear Admiral Stephenson wrote Dr. Lewis H. Weed, Chairman of the Division of Medical Sciences, National Research Council, under date of Dec. 2, 1942, requesting that a suitable committee of pathologists be appointed to receive pathological material from the Commission and to cooperate in the study of the disease so that results might be available for an early report. Dr. Weed decided that there might be a broader field for a Committee on Pathology and on Jan. 5, 1943, wrote the Surgeons General of the Army and Navy suggesting that such a committee be formed. On prompt receipt of favorable responses from these officers, Dr. Weed appointed as organizing members of the Committee Dr. Paul R. Cannon, Dr. Robert A. Moore, Dr. Alwin Pappenheimer, Dr. S. Burt Wolbach, and Dr. Howard T. Karsner, Chairman. The Committee was subsequently enlarged and now is comprised of 13 members.

The first meeting was held Feb. 16, 1943. At that meeting and at no less than eight subsequent meetings the request of Rear Admiral Stephenson was the principal item of the agenda. Lieutenant Commander W. B. McAllister (MC), U. S. N. R., was the pathologist of the Commission and went to Cairo with the first group of the Forward Echelon. He was to perform the autopsies, prepare protocols, make tissue blocks in fixatives recommended by the Committee, and forward the wet blocks to the Army Medical Museum for processing and distribution to assigned members of the Committee. The entire material was to be combined in a joint report of Lieutenant Commander McAllister and the Committee. This was confirmed at subsequent meetings of the Committee. Although Captain Cushing assured the Committee on May 26, 1943, that the material would soon be sent from Cairo, unavoidable delays deferred the receipt until 1944.

Brigadier General Leon Fox and Brigadier General Bayne-Jones attended a meeting of the Committee on June 2, 1944, and stated that material from 23 cases had been forwarded. Sections were made at the Army Medical Museum and sent to the assigned members of the Committee. Reports, amply illustrated by photomicrographs, were prepared on cardiovascular system by Dr. Howard T. Karsner, on respiratory tract by Dr. A. M. Pappenheimer, on central nervous system by Dr. R. D. Lillie, and on the other material by Dr. Robert A. Moore. These were presented at a meeting on March 31, 1945, collated, and forwarded via the Chairman of the Division of Medical Sciences to the Director of the Commission.

Under date of Jan. 25, 1949, Lieutenant Commander McAllister issued Report No. 1 on "The Pathology of Louse-Borne Typhus Fever from the Epidemic of 1943-1945 in Egypt" from the Naval Medical Research Institute, Bethesda, Md., Project NM 007 017 (X696). By the time he began work at the Institute he had collected material from 39 proved cases, including the material from 23 cases sent earlier to the Committee. This was evidently not a final report, but further work by Dr. McAllister was interrupted. When the Committee met May 10, 1951, Dr. McAllister presented a somewhat amplified report. By this time the Armed Forces Institute of Pathology had prepared sections on all 41 cases, and these were assigned

TABLE 1.—*Important Clinical and*

Autopsy No.	Case	Age, Yr.	Sex	Total Duration, Days	Admission Day	Highest Temp., F.	Chills	Laboratory: Weil-Felix Reaction, 1:	Laboratory: Complement Fixation (Typhus)	Laboratory: Virus Isolated	Circulatory System: Highest Pulse	Circulatory System: Heart	Circulatory System: Blood Pressure	Circulatory System: Cyanosis	Circulatory System: Shock	Circulatory System: Blood Vol.
3	C	20	M	11	10	36.0	..	250	..	..	74	..	.....	..	+	..
4	C	25	M	7	3	39.8	..	...	..	..	112	N	.....	..	..	..
5	C	32	M	13	8	39.0	..	O	..	..	125	N	.....	..	..	..
6	C	30	F	9	5	38.6	..	250	..	..	136	N	.....	..	..	..
7	C	40	M	11	5	39.2	+	O	..	..	120	N	.....	..	..	..
8	C	20	F	14+	..	39.2	..	250	..	..	130	N	.....	..	..	..
9	T	55	M	14	8	37.8	+	640	+	+	96	N SM	96/58	O	O	..
11	T	25	M	14	10	40.0	+	O	—	..	140	N	114/70	O	O	..
12	C	35	F	13	8	39.5	+	250	..	..	108	N	.....	..	..	..
13	C	35	M	15	4	40.0	..	250	..	..	130	W	.....	..	..	..
14	C	20	M	14+	..	37.5	..	250	..	..	120	N	.....	..	..	..
16	C	30	M	9	8	37.0	+	O	..	..	100	N	.....	..	..	..
17	C	28	F	14	10	39.0	..	O	..	..	130	W Dy	.....	..	+	..
18	C	50	F	13	8	40.0	+	O	..	..	130	W	.....	..	..	..
19	C	40	M	..	..	....	O	...	..	..	....	..	.....	..	..	..
20	C	49	F	15	10	40.0	+	...	..	..	....	N	.....	..	..	..
21	C	18	M	18	8	39.5	O	250	..	..	130	N	.....	..	..	..
22	T	21	M	17	7	40.2	+	2560	..	+	150	N I	106/66 80/40	O	O	..
23	C	30	M	6	4	39.5	..	250	..	..	130	N	.....	..	..	..
24	C	10	M	..	..	....	..	...	..	..	....	..	.....	..	..	..
26	C	25	M	..	..	39.5	..	250	..	..	....	..	.....	..	..	..
27	C	25	M	..	..	....	..	....	..	..	....	..	.....	..	..	..
28	T	38	M	11	8	40.3	O	...	..	..	135	N G P	98/70	O	+	..
30	T	36	M	16	5	40.8	O	+	..	..	135	N G	125/65 100/60 120/70	O +	O	..
33	T	35	M	12	8	40.8	O	640	..	..	140	N G	96/60 76/20	O	O	..
34	T	70	M	10	2	40.7	O +	—	—	..	128	N	106/72 84/50	O +	+	..
35	T	30	M	11	4	40.7	O	+	..	..	140	N	118/64 90/65 60/40	O	O	..
37	T	43	M	13	6	40.5	O	...	..	..	128	N VE	100/50	O +	O	..
38	C	35	M	6	4	40.0	O	...	..	..	....	N W	.....	..	..	..
39	T	40	M	7	5	40.3	O	...	..	..	142	G	94/60 66/44	O	O +	N
40	T	45	M	12	5	40.8	+	+	O	..	140	N	120/80 90/54	O +	O	N
41	T	48	M	16	7	40.2	+	320	—	..	144	N	72/50 108/60 98/50	O +	O	N
42	..	..	..	..	..	....	..	...	..	..	....	..	.....	..	..	..
43	C	30	M	16	12	39.4	O	...	..	..	....	N W	.....	..	..	..
44	..	23	M	12	5	40.9	..	O	..	..	148	..	.....	..	..	..
45	C	50	M	6	3	40.6	+	...	..	..	....	N	.....	..	..	..
46	T	18	M	14	9	40.8	+	20	+	..	140	N G	122/80 86/50 60/40	O +	O	100
47	..	41	M	16	7	....	..	...	..	..	....	..	.....	..	..	..
49	T	18	M	16	8	40.6	O	5120	+	..	140	N	70/60 90/50	O	+	..

* Key to abbreviations: A, albumin; Ca., casts; C, Cordon case; Cl, clear; Dl, delirious; Du, dull; Dy, dyspnea; D, deaf; G, gallop rhythm; I, irregular; N, normal; O, absent; Ol, oliguria; P, premature beat; R, lower lobe of right lung; SM, systolic murmur; Tw, twitching; T, Typhus Commission case; U, unconscious; VE, ventricular extrasystoles; V, vomiting; W, weak. Progression of signs is indicated by the vertical position of the values. All patients had a rash and a typical clinical course.

*Laboratory Findings in Cairo Cases**

Kidneys					Nervous System			Respiratory System			Alimentary System			
Output	N. P. N., Mg./100 Cc.	Renal Clearance, % of Normal	Specific Gravity; Misc.	Dehydration	Headache	Mental State	Reflexes	Moisture	Consolidation	Rate	Diarrhea	Hepatomegaly	Splenomegaly	Tympanites
..	..	..	.....	+	..	U	..	+	O	..	..	..	..	..
..	..	..	.....	+	+	U	..	+	O	..	..	..	..	..
..	..	..	.....	..	+	..	..	+	O	..	..	..	+	..
N	..	..	.....	..	..	U	..	+	O	..	..	..	..	..
..	..	..	.....	..	..	U	..	O	O	..	..	O	O	..
..	..	..	.....	..	..	U	..	O +	O	..	..	O	+	..
N	181	..	.....	O	+	Dl	N	+	O	..	O	O	O	+
N	90	..	1.024 A.	+	+	Dl	N	O	O	22	O	O	O	O
..	..	..	.....	..	+	..	..	+	O	..	..	..	..	+
..	..	..	.....	..	+	U	..	O +	O	..	..	..	..	..
..	..	..	.....	..	..	U	..	+	O	..	+	..	..	..
..	..	..	.....	..	+V	..	..	+	O	..	..	..	O	..
..	..	..	.....	+	..	Dl	..	O	O	..	..	..	..	..
..	..	..	.....	..	+	U	..	..	..	..	..	..	..	+
..	..	..	.....	+	..	..	..	..	..	..	..	O	O	..
..	..	..	.....	..	..	..	..	O	O	..	..	O	O	..
..	..	..	.....	+	..	..	..	O	O	..	..	O	O	..
N	96 69	..	1.030	+	+V	Dl U	N	O +	O R	40 54	O	O	+	O
..	..	..	.....	..	+	..	..	O	O	..	..	..	..	..
..	..	..	.....	..	..	..	..	..	..	..	..	..	..	..
..	..	..	.....	..	..	U	..	+	O	..	..	..	..	..
..	..	..	.....	..	..	..	..	..	..	..	..	..	..	..
Ol	90	..	1.012 A. Ca.	O	+V	Dl	N Tw	O +	O	56	+	O	+	O +
Ol	30 75	..	1.015 A. Ca.	O	+	Cl U	O	+	O +	34 54	O	O	O +	O +
N	75	137	1.015 A.	+	+	Du	N	O +	O +	36 40	O	O	+	O
N	67	110 56	1.015	+ O	O	Du	N D	O +	O	32 38	O	O	O	+
Ol	30 110	190 10	1.023 A.	O	+	Cl Dl	N	+	O	36	O	O	+	O
Ol	30 75	110 20	1.015 A. Ca.	O	+	Cl Dl	D N	O +	O	40	O	O	+	O
..	..	..	.....	..	..	Du		+	..	..	..	..	+	..
Ol	130	20	1.025 A. Ca.	O	+	Dl	N	+	O	36 40	+	O	+	+
N Ol	34 175	100 10	1.020 A. Ca.	O	+V	Dl	N	+	O	22	+	O	O	+
N	68 90 52	33	1.017 A. Ca.	O	+	Cl Dl	N	+	+	32	O	+	+	O
..	..	..	.....	..	..	..	..	..	..	..	..	..	..	..
..	..	..	.....	..	+	..	..	+	..	..	..	..	+	..
..	..	..	.....	..	..	..	..	..	..	..	..	..	..	..
..	..	..	.....	..	..	..	..	O	..	..	..	..	O	..
N	43 82	20	1.018 A. Ca.	O	O	Dl U	N	+	O	44	O	O	O	O
..	..	..	.....	..	..	..	..	..	..	..	..	..	..	..
N	108 149 41	..	1.010 A. Ca.	O	+V	Cl	N	O	O	38	+	O	+	+

TABLE 2.—*Summary of Organ Incidence*

Autopsy No.	Skin	Myocardium	Endocardium	Epicardium	Aorta	Trachea	Lungs	Tongue	Tonsil	Palate and Oropharynx	Salivary Glands	Esophagus	Stomach	Intestine	Colon	Liver	Gall Bladder	Pancreas	Kidney	Urinary Bladder	Uterus	Ovary	Vagina	Testis	Epididymis	Sperm Duct	Prostate
3	..	1,3	0	3	..	..	0	1	0	..	..	0	0	1	0	0	1	0	1,3	..	..	..	..	..	..	..	1
4	..	1,3	0	0	..	..	0	4	1	..	0	..	0	0	0	0	..	0	1,3	0	..	..	..	..	..	..	0
5	1	1,3	0	0	..	..	1	4	..	..	0	..	1,2	1,2	..	0	0	1	1,3	0	..	..	..	..	..	..	..
6	1,2	1,3	0	0	..	..	0	..	..	..	..	..	0	0	..	0	0	0	1,3	..	0	0	0	..	..	..	..
7	1,2	1,3	3	1	..	..	0	4	0	..	..	..	0	..	0	0	..	0	1,3	0	..	..	..	..	..	..	..
8	1	1	0	1	..	..	0	0	..	..	0	..	0	..	0	0	..	0	1,3	..	0	0	..	..	..	..	..
9	1	3	0	0	0	..	0	4	0	..	0	..	0	0	0	0	..	0	1,3	1	..	..	..	0	..	..	0
11	1	3	3	0	..	..	1	4	..	..	..	..	0	..	0	0	..	0	1,3	..	..	..	..	1,3	..	0	0
12	1	1	0	0	..	..	0	0	..	1	1	..	0	..	0	0	..	0	1,3	1	0	0	..	..	..	..	..
13	0	1,3	0	3	..	..	0	4	..	..	..	..	0	0	..	0	0	1	1,3	0	..	..	..	..	..	0	0
14	..	0	0	3	..	..	0	..	..	..	..	..	..	1,2	0	0	..	0	1,3	0	..	..	..	..	..	..	0
16	0	3	0	0	..	..	..	4	0	1	0	..	0	0	..	1,2	0	0	1,3	0	..	..	..	1,3	0	1	1
17	0	1	0	0	..	..	0	0	..	..	..	..	0	..	0	0	0	0	1,3	0	0	0	..	..	..	..	..
18	1,2	3	3	0	..	..	0	4	..	1,2	0	1,2	0	0	..	0	..	0	1,3	0	0	1	1	..	..	..	..
19	1	3	0	0	0	..	0	4	0	..	..	..	0	0	0	0	0	0	1,3	0	..	..	..	0	0	..	0
20	1	3	3	0	..	..	0	4	0	..	0	..	0	0	..	0	0	0	1	0	0	0	..	..	..	..	..
21	1	1,3	0	0	..	..	0	4	0	..	..	0	..	0	0	0	..	0	1,3	..	..	..	..	..	..	..	0
22	1,2	4	3	4	..	..	0	0	0	..	..	..	..	0	..	0	0	1	1,3	0	..	..	..	1,3	0	..	0
23	1,2	3	3	0	3	0	1	4	..	..	..	1,2	0	..	..	1,2	..	0	1,3	1	..	..	..	1,3	..	..	0
24	1	3	0	0	..	0	0	0	..	..	0	..	0	0	..	0	0	0	1,3	..	..	..	..	0	..	0	..
26	1,2	3	0	0	2,3	..	0	4	0	0	0	..	1,2	0	..	0	0	0	1,3	..	..	..	..	..	..	..	0
27	..	3	0	0	..	..	0	..	..	..	..	..	..	0	0	0	..	0	1,3	..	..	..	..	..	..	..	0
28	..	3	0	0	..	..	0	..	0	..	..	..	..	..	..	0	0	..	3	0	..	..	..	1,3	..	..	..
30	..	4	3	3	..	..	..	..	1,2	..	0	..	1	..	..	..	..	..	..	..	..	..	..	1,3	..	..	0
33	1	1,3	3	0	..	..	0	4	..	..	..	..	1	0	0	0	..	..	1,3	..	..	..	..	..	..	..	0
34	..	3	0	0	..	0	0	..	..	..	..	..	..	..	0	..	..	..	..	..	..	..	..	..	..	..	..
35	..	..	..	..	..	..	..	..	..	..	..	..	0	0	..	0	..	..	1,3	0	..	..	..	..	..	..	..
37	..	0	0	0	..	..	0	..	..	..	..	..	..	0	..	0	..	..	1,3	..	..	..	..	..	..	..	..
38	..	3	0	0	0	..	0	4	..	..	..	..	..	0	..	..	0	0	1	0	..	..	..	3	..	0	0
39	0	3	0	0	0	0	0	1	..	..	0	..	..	0	0	0	..	0	1,3	0	..	..	..	1,3	..	..	..
40	..	3	0	0	..	..	0	4	0	1,2	0	0	0	0	0	0	..	0	0	..	..	..	..	0	..	..	..
41	1	1,3	3	3	0	..	0	..	..	..	..	..	0	0	..	0	..	0	1,3	..	..	..	..	4	..	..	1
42	1,2	3	3	3	3	..	0	4	0	..	..	1	..	..	..	..	..	..	1,3	..	..	..	..	1,3	..	..	2
43	1,2	1,3	3	0	0	..	0	4	..	..	..	..	1,2	..	0	0	..	0	1,3	..	..	..	..	1,3	..	..	..
44	1,2	..	..	..	0	1	0	4	0	..	0	..	..	..	..	0	0	0	1,3	0	..	..	..	..	..	..	0
45	1,2	1,3	0	0	..	..	0	..	..	..	..	..	..	..	..	..	..	..	1	..	..	..	..	..	..	..	..
46	..	3	3	0	..	..	0	4	..	1,2	0	..	0	..	..	0	..	..	1,3	..	..	..	..	..	..	..	0
47	..	3	0	0	..	..	0	..	..	..	..	..	0	0	0	0	..	0	1,3	..	..	..	..	..	..	..	0
49	1	3	0	0	..	..	1	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	1,3	..	..	..
Total Cases																											
39	26	37	37	37	10	5	36	27	15	6	15	6	26	24	18	33	14	29	36	19	6	6	2	16	3	5	21
No. Cases Involved														3	0	2	1	3	34	3	0	1	1	12	0	1	4
22	25	12	9	3	1	4	22	2	5	1	3	5															

* Key to symbols: 1, vascular lesions; 2, nodular lesions; 3, diffuse cellular accumulations; 4, presence of all 3 lesions; 0, absence of typhus lesions; G+, Gram-positive cocci; G—, Gram-negative cocci; blank space, no tissue available.

*of Typhus Lesions in Cairo Cases**

Seminal Vesicles	Spleen	Lymph Nodes	Bone Marrow	Adrenal	Pituitary	Thyroid	Thymus	Muscle	Parathyroid	Bone	Brain	Spinal Cord	Meninges	Spinal Nerves	Rickettsia-Like Bodies in Lungs	Coccl in Lungs	Complications†
0	0	..	..	0	..	..	..	..	..	..	1,2	..	3	..	4+	G—	Hem. brpn., r. l., l. l.
..	..	0	..	0	..	0	..	0	0	..	1,2	2	3	..	0	G+	Brpn., r. l., l. l.
0	0	0	0	1	..	0	..	1	..	..	1,2	1	..	..	4+	G+	Brpn., r. l., l. l.
0	..	..	0	0	..	0	..	..	..	..	1,2	..	..	..	4+	0	Pulmonary aspergillosis, r. l., l. l.
..	..	0	0	1	..	0	..	..	..	..	2	2	..	..	2+	G—	Brpn.; cal. schis. ova, bladder
..	0	..	0	0	..	0	..	..	..	..	2	1,2	..	..	0	G+	Brpn., r. l., l. l.; decubiti; necrosis, skin of feet; enteritis
..	0	0	..	3	..	0	0	..	..	..	2	2	..	2	1+	0	Brpn., l. l.
0	0	..	0	1	..	0	..	1	..	..	1,2	..	3	..	0	....	Inter. pn.; ac. colitis
..	0	..	0	1	..	..	0	..	..	..	1,2	2	..	..	0	G+	Brpn., r. l., l. l.; ac. colitis; diffuse cirrhosis; decubiti
0	0	..	0	1	..	0	0	..	..	..	1,2	1,2	..	2	0	....	Hem. brpn., r. l., l. l.; cal. schis. ova, prostate; ac. colitis
..	0	..	0	0	..	0	..	0	..	..	2	0	3	..	....	....	L. pn., r. m., r. l., l. l.; ac. meningitis; pericarditis; schis., bladder, lung
..	0	..	0	3	..	1	0	2	..	..	1,2	1,2	3	..	....	....	Schis., kidney, testis
..	0	..	0	0	..	0	..	0	..	..	2	0	..	..	0	0	Brpn.; schis., bladder
..	0	..	0	2	..	0	..	..	..	..	0	0	0	..	0	0	Inter. pn.
..	0	0	0	0	..	..	0	..	..	..	2	0	..	..	1+	0	Brpn., l. u., l. l., r. l.; schis., bladder, seminal vesicles
..	0	..	0	0	..	0	0	1	..	..	2	0	..	..	0	0	Inter. pn., r. l., l. l.; diffuse cirrhosis; schis., bladder; ac. colitis
..	0	..	0	0	0	..	..	..	..	..	1,2	1,2	3	..	3+	0	Brpn., r. l., l. l.; uremic frost; ac. colitis; schis., sem. ves.
..	0	0	1	1	2	0	..	..	0	..	1,2	2	..	..	4+	G+	Brpn., all lobes
0	0	0	0	0	1,2	..	..	0	..	..	2	2	..	..	0	....	Inter. pn., r. l., l. l.; schis., lungs, bladder, sem. ves.
..	0	..	0	..	..	0	..	0	..	..	2	2	..	..	0	0	Inter. pn., r. l., l. l.
0	0	..	0	0	2	0	..	1,2	..	..	1,2	2	..	..	3+	0	Inter. pn., upper and lower lobes; schis., lungs
..	0	..	0	0	..	..	..	..	..	..	2	0	..	..	0	0	Brpn., r. l., l. l.; inter. pn.
..	0	..	0	..	..	0	..	0	..	..	1,2	2	..	..	0	....	Schis., liver; ascites (3,000 cc.); anasarca
..	..	..	0	..	0	..	..	0	..	..	1,2	2	..	..	....	....	Schis., prostate cal.
..	0	0	0	1,2	..	0	..	..	..	..	1,2	1,2	..	..	1+	0	Inter. pn.
..	..	..	..	..	..	..	..	..	..	..	0	..	..	..	0	0	Brpn.; schis., colon, lung
..	0	..	..	..	..	..	..	..	..	..	0	0	..	..	....	....	Schis., liver, stomach, intestine, bladder
..	..	..	..	..	..	..	..	..	..	..	2	0	..	..	0	0	Diffuse pneumonitis; schis., liver, lungs
0	0	..	0	0	..	..	0	..	..	..	..	..	..	..	4+	0	Brpn.; schis., lung, prostate, kidney, bladder, vas., sem. ves.
..	0	..	0	0	..	0	..	0	..	..	..	0	..	..	0	0	Brpn.; schis., colon, bladder, lung, liver
..	..	..	0	0	..	..	0	0	..	..	..	..	..	..	3+	0	Brpn.
..	0	..	0	0	0	..	..	..	..	..	..	..	..	..	0	0	Bronchiectasis
1	..	..	0	2	..	..	..	..	..	..	..	..	..	..	1+	0	Inter. pn.
..	0	..	0	0	..	..	..	0	..	..	..	..	..	..	....	....	Purulent bronchitis and colitis
..	0	..	..	0	..	0	..	..	..	1	..	..	..	..	0	0	.....
..	..	..	0	..	..	..	..	..	..	..	..	..	..	..	0	G+	Brpn.
..	0	..	0	0	..	..	..	1,2	..	..	..	..	..	..	3+	G—	Hem. brpn.
..	0	..	0	0	..	..	..	..	..	..	..	..	..	..	0	0	Brpn.; schis., prostate
..	0	..	..	..	..	..	..	..	..	..	1,2	1,2	..	..	....	....	Brpn.; schis., lung
9	30	8	31	31	6	20	8	16	2	1	29	26	7	26	33	28	
1	0	0	1	10	3	1	0	6	0	1	26	17	6	2	14	9	

† Ac., acute; brpn., bronchopneumonia; cal., calcified; hem. brpn., hemorrhagic bronchopneumonia; inter. pn., interstitial pneumonia; l. l., lower lobe of left lung; l. pn., lobar pneumonia; l. u., upper lobe of left lung; r. l., lower lobe of right lung; r. m., middle lobe of right lung; schis., schistosomiasis; sem. ves., seminal vesicles.

for study to the list of contributors to this monograph. Gross material of central nervous system was available from some of the latter cases and was sufficient to justify a survey of topography of lesions, and this was undertaken by Lieutenant Commander Boyd Black and Dr. David E. Smith. Dr. Andrew Yeomans, who was active in the clinical studies in Cairo, consented to collaborate on certain clinical aspects of the disease. Dr. Bayne-Jones had been authorized by President Truman to close out the affairs of the Commission and sent back to the Committee the reports and photomicrographs made by the original list of investigators. These were reviewed and supplemented by earlier authors. The manuscripts and photomicrographs furnished by the contributors more recently assigned were added, and Dr. William B. Wartman agreed to act in an editorial capacity to condense the text, select a limited number of illustrations for publication, and correlate the findings with those reported previously in the literature.

At a meeting of the Committee, Nov. 1, 1952, Dr. Wartman's editorial arrangement was considered in detail and, with a few minor alterations, was approved. Decision to publish was reached because this report differs in certain measure from others which are quoted below. It is the joint effort of a considerable number of pathologists with differing experience and background. It offers a topographic survey of lesions of the central nervous system. It introduces certain new features of some of the cases of pneumonia suggestive of rickettsial inflammation. It appears to settle the lack of significance of dehydration in the lesions of the disease. It may well be the last study of the pathology of typhus not influenced by the use of antibiotics.

MATERIAL AND METHODS

WILLIAM B. WARTMAN, M.D.

THIS ACCOUNT of the pathology of epidemic typhus is based on material obtained from 39 fatal cases that were studied by the United States of America Typhus Commission in 1943 to 1945 during an epidemic of the disease in Cairo, Egypt. The pertinent clinical and laboratory data were abstracted from the patients' charts and are summarized in Table 1 and the important pathological findings in Table 2.

There were two groups of patients. Group 1 consisted of 14 patients who were cared for by Lieutenant Commander Andrew Yeomans (MC), U. S. N. R., in a special ward assigned to the Typhus Commission. Careful histories and physical examinations were obtained, and in many cases special studies were made of blood electrolytes and of cardiac and renal function. The patients were given supportive treatment, adequate amounts of fluid either by mouth or by vein, and sulfonamides or penicillin when secondary bacterial infection, such as pneumonia, occurred. The second group of patients were cared for on the ordinary fever wards of the Cordon Hospital; the clinical studies were not as detailed as those of the patients in the first group, and treatment was entirely supportive. With the exception of two patients on the Typhus Commission Ward who were treated with paraaminobenzoic acid, none had antirickettsial drugs or vaccinations, since these were not available at the time. The paraaminobenzoic acid had no demonstrable effect upon the course of the two patients. The records of Cases 30 and 35 have been previously published in a study of the development of azotemia (Yeomans, Snyder, Murray, Ecke, and Zarafonetis, 1945) and the records of Cases 40, 41, and 49 in a report on the

metabolic disturbances in typhus (Tierney and Yeomans, 1946). The material from Cases 3 to 26, inclusive, was used by Allen and Spitz (1945) in their study of the comparative pathology of scrub typhus, epidemic typhus, and Rocky Mountain spotted fever.

One of the objectives of the Typhus Commission was to compare the clinical and pathological manifestations of the disease in these two groups of patients. This paper is concerned primarily with the pathological findings. No significant differences were observed in the lesions in the two groups. Secondary bacterial infection, for unknown reasons, was not prevalent in either group, and it therefore seems reasonable to attribute the lesions to the effects of the rickettsiae alone. Certainly this seems justifiable in the patients who were cared for on the Typhus Commission

TABLE 3.—*Summary of Important Complications in Cairo Cases*

Complication	No. of Cases	
	Examined	Involved
Bronchopneumonia	36	21
Interstitial pneumonia	36	10
Lobar pneumonia	36	2
Bronchiectasis	36	4
Schistosomiasis	40	19
Urinary bladder	19	9
Lung	36	8
Prostate	21	4
Seminal vesicles	9	3
Colon	18	3
Liver	33	3
Kidney	36	2
Testis	16	1
Vas deferens	5	1
Stomach	26	1
Pancreas	29	1
Ascariasis	40	1
Sundry infections	40	17
Acute colitis and proctitis	18	7
Acute enteritis	25	1
Acute prostatitis	21	2
Acute meningitis	..	1
Chronic pyelonephritis	36	2
Syphilitic aortitis	10	3
Cirrhosis (diffuse)	33	2

Ward, for they were treated with sulfonamides and penicillin as soon as secondary bacterial infection was detected. Thus the findings in the Cairo cases are of value for comparison with other reported epidemics in which such secondary bacterial complications have been both severe and common.

Detailed reports of the pathology of epidemic typhus have been written by Ceelen (1919) and by Wolbach, Todd, and Palfrey (1922). In recent years the pathology of the disease has been studied during epidemics in Russia (Abrikosov, 1941), Austria (Chiari, 1943), Roumania (Danielopolu and Lupu, 1936), Brazil (Fialho, 1933), Guatemala (Golden, 1945), Chile (Herzog, 1935), and in the civilian population of Soviet territory occupied by the Germans during World War II (Randerath, 1943; Schopper, 1943). Wolbach (1948, 1950) has written succinct and thoughtful accounts of the general pathology of human rickettsial diseases in the light of modern knowledge of their pathogenesis. Allen and Spitz (1945)

have made a study of the comparative pathology of scrub typhus (American military personnel in the Southwest Pacific), epidemic typhus (Egypt, 1943), and Rocky Mountain spotted fever (United States).

Complete autopsies were performed on the Cairo patients immediately after death by Lieutenant Commander W. B. McAllister (MC), U. S. N. R. Blocks of tissue were taken from all organs and fixed in Regaud's fluid and 4% formaldehyde solution. The wet tissues were sent to the Armed Forces Institute of Pathology, Washington, D. C., where they were embedded in paraffin, and histological sections were cut and stained with hematoxylin and eosin. Special histological, bacterial, and rickettsial stains were prepared as indicated. The whole brains were fixed in formalin, and, after fixation, blocks were embedded in both paraffin and celloidin for histological and topographical studies. Each of the authors of this report examined a complete set of the slides and wrote a detailed account of the findings in the organs assigned to him. These accounts were then reviewed by the other authors, as well as the members of the Committee on Pathology of the National Research Council, so that the final report expresses the opinions of a group of pathologists rather than of individuals. In presenting the findings, a brief summary of the general pathology of epidemic typhus is given, followed by detailed discussions of the lesions in the various organs of the body. In these discussions the general plan has been first to give a résumé of the important facts about the lesions in the organs under discussion; second, the findings in the Cairo cases, and third, a comment on these findings in the light of the literature published since the monograph of Wolbach, Todd, and Palfrey (1922). Thus the reader can find all the known facts about the special pathology of each organ in one place. At the end of the report there is a discussion of some of the problems of general interest, most of which still wait solution.

BRIEF SUMMARY OF PATHOLOGY OF EPIDEMIC TYPHUS

IT IS PROBABLE that in epidemic typhus the rickettsiae first localize and multiply in endothelial cells of the intima and then enter the large mononuclear cells that accumulate about blood vessels (Wolbach, 1950). The first reaction consists of proliferation of vascular endothelium and of mononuclear cells around blood vessels, often resulting in nodular accumulations. When the disease is severe, necrosis of arterial walls may be conspicuous, but in milder cases the media is not markedly involved. As a result of the intimal involvement, thrombosis occurs early. Hemorrhages may be prominent. This focal, proliferative, endothelial, and infiltrative response to infection is present in skin, brain, heart, skeletal muscles, spleen, adrenals, and other organs in varying degree and distribution and gives rise to some of the clinical signs. The skin is invariably affected as shown clinically by the characteristic rash. In the central nervous system the focal lesions, which are of microscopic size, are referred to as Fraenkel nodules and are of the same origin as the cutaneous lesions. The myocardium is commonly affected. The lungs may show bronchopneumonia, but in the past this has usually been attributed to secondary bacterial infection rather than to rickettsiae. Although renal failure may dominate the clinical course, particularly in severely ill patients, exact clinicopathologic correlation is often not possible.

SKIN AND SUBCUTANEOUS TISSUES

ZOLA K. COOPER, M.D.

RÉSUMÉ

SKIN lesions are practically constant in typhus, although multiple sections may have to be examined before they are discovered. The characteristic changes are a combination of vascular lesions and perivascular accumulations of mononuclear cells. The vascular lesions consist of swelling and proliferation of the endothelium, thrombi, acute inflammation, and necrosis. The capillaries are especially involved, often by the fifth day. These lesions are nodular, but, in addition, there may be diffuse perivascular infiltrations of mononuclear phagocytes, lymphocytes, plasma cells, and mast cells with a rare polymorphonuclear neutrophile. Hemorrhages may also be found in the corium.

CAIRO CASES

A macular rash was always present, being most evident on the thorax, abdomen, thighs, and upper arms. It was less conspicuous on the forearms and lower legs, and least so on the hands, feet, head, and neck. The palms and soles were spared. The individual macules were reddish brown, irregular in outline, and pale at the edge. However, many early lesions were red and did not show peripheral pallor. In patients who died after a long illness, the rash was brown and not easily seen. When desquamation began, usually during the third week of fever, the rash faded rapidly and was commonly invisible at the time of autopsy, although slightly indurated and elevated nodules were usually palpable. In 40% of early cases, the macules were so numerous over the pectoral, deltoid, and scapular regions that they became confluent and formed large irregular discolored areas accompanied frequently by scattered petechiae.

A second cutaneous manifestation of typhus, which was observed particularly in patients who died before the third week of fever, was a diffuse livid bluish red subcuticular mottling of the face and neck that gradually faded away at the level of the nipples. This mottling, coupled with puffiness of the face, produced a characteristic facies.

Uremic frost was present in Case 21.

Sections of skin from 26 patients were examined microscopically. The most distinctive changes occurred in the dermis. Slight to moderate edema was pronounced in the papillary portion and less so elsewhere. The blood vessels were dilated, especially those in the upper dermis, and many were engorged. In a majority of patients, the endothelial cells lining the blood vessels were swollen and the walls of some of the large vessels both in the dermis and the subcutaneous fat were thickened. In 22 of the 26 patients vascular lesions were present in capillaries. Large arteries and veins in the deeper corium and occasionally in the hypodermis showed subendothelial infiltrations of various mononuclear cells. Polymorphonuclear leucocytes about thrombi were observed. No areas of hemorrhage were found in the dermis, but in Case 42 there were small hemorrhages in the subcutaneous fat. Perivascular and perifollicular infiltration of mononuclear cells, lymphocytes, and occasional plasma cells was present in every instance. The density and composition of the infiltrate varied and in some instances was only slight. In

one there was necrosis of the lower dermis and subcutaneous fat. There was no evidence of basophilic degeneration of collagen. In many instances there appeared to be an excess of mast cells, particularly near the junction of dermis and hypodermis.

The changes in the epidermis were less specific than those in the dermis. The epidermis was normally thick in most instances or slightly atrophic. In no case was there acanthosis, but hyperkeratosis was constant. Hair follicles and sweat gland ducts were distended and plugged with keratin. The granular layer was very thin in most instances so that only a few cells containing keratohyalin granules were found, and they were rather patchily distributed. In six patients there was edema of the epidermis, and it was for the most part intracellular. Liquefaction degeneration of the basal layer was present in two instances. The pigment in the basal layer was normal for dark-skinned people.

Changes were present in the sweat glands in about half of the patients. The mouth of the sweat duct was dilated and filled with keratin, and the upper part of the duct was enlarged. In the secretory portion of the gland the cells were swollen and vacuolated. This was probably an incidental finding and might be related to the fact that these persons had had fever.

COMMENT ON LESIONS OF SKIN AND SUBCUTANEOUS TISSUES

The characteristic rash of typhus has been well described by Wolbach, Todd, and Palfrey (1922) and more recently by Yeomans (1948) and others. It appears first on the trunk, later spreads to the extremities, and almost always spares the face and scalp. Rarely the palms and soles are affected. Usually it appears between the fifth and seventh days and occasionally as early as the fourth day of disease. The eruption often develops rapidly within a few hours; at first the lesions are faint rose-colored macules of irregular shape that fade on pressure, but after 24 to 48 hours many of them are dark red and indurated and will not fade on pressure. At the height of the eruption, which is usually about the eighth day, the entire body may be covered with maculopapular lesions. In some instances petechiae may involve hair follicles. An early papular rash is said to indicate a severe infection, although fulminating cases are described in which no rash is ever detected. Wolbach, Todd, and Palfrey (1922) examined the lesions in living patients with the aid of the skin microscope and found that each consisted of "a tangle of dark red blood vessels."

The characteristic combination of vascular lesions and perivascular accumulations can usually be detected on microscopic examination after the fourth day. Wolbach, Todd, and Palfrey (1922), using both biopsy and autopsy material, studied the pathogenesis of the skin lesions in the Warsaw cases. The earliest changes were seen in specimens taken for biopsy on the first day of the rash and consisted of swelling of the endothelium of capillaries, small arteries, and veins. The swollen cells often occluded the capillaries and frequently were in mitosis. They contained minute coccoid bodies thought to be rickettsiae, but this was not proved. Polymorphonuclear leucocytes were abundant in the perivascular tissues. By the fifth day of disease mural and occlusive thrombi were found in arteries and veins in the lower layers of the corium, and distinct nodules were present. Some thrombi were accompanied by very little perivascular reaction. After the fifth day there was further development of the nodules and involvement of large vessels in the

deepest layers of the corium and in the subcutaneous fat. On and after the eighth day hemorrhages were found about blood vessels in the corium and were attributed to necrosis of endothelium dependent partly on arterial thrombosis and partly on venous stasis. Even as late as the 9th to 11th days acute lesions of the intima without marked perivascular reaction were found in capillaries, small arteries, and veins. Repair of the thrombi occurred by organization starting as early as the 15th day. Repair of nodules was more difficult to follow but probably occurred by granulation tissue and fibrosis. Proliferation of fibroblasts at the periphery of the nodules occurred on the 18th day and partial disappearance of infiltrative cells on the 20th to 24th days. Lymphocytes, plasma cells, and mast cells increased with the age of the nodule, as did phagocytosis. According to Randerath (1943), the exanthema may be detected microscopically for as long as 27 days. Allen and Spitz (1945) have described disintegration into chromatin dust of the nuclei of swollen endothelial cells and the development of "infarct-like hemorrhagic suppurative necrosis of portions of the corium in association with severe arteritis." They also observed focal spongiosis, patchy parakeratosis, and focal liquefaction degeneration of the basal layers of the epidermis. Lymphangiectasia in the corium has been described (Golden, 1945).

Necrosis of skin and subcutaneous tissues has been described rather commonly. It has been attributed to extension of thrombi from the superficial arteries to those deep in the skin and subcutaneous tissues following centripetal extension of the infection (Wolbach, Todd, and Palfrey, 1922). This process is probably facilitated by a moderate degree of stasis and, of course, inadequate nursing care. Mild lesions of the skin are not uncommon (Killian and Obertreis, 1943; Sylla, 1942), but massive necrosis involving even muscles is also described (Wolbach, Todd, and Palfrey, 1922; Killian and Obertreis, 1943; Randerath, 1943). It is particularly prone to occur over bony prominences, and Sylla (1942) observed that it usually began from the 11th to the 13th day and was commonest in cold weather or following exposure.

Erysipelas, carbuncles, and multiple abscesses have also been reported, but these are probably the result of secondary infection (Killian and Obertreis, 1943).

Walther (1942) has described patients who suffered a recurrence of slight fever accompanied by urticaria during convalescence. He attributed this to the development of an allergic state. Others have suggested that it is not related to typhus but to some therapeutic agent.

CARDIOVASCULAR SYSTEM

HOWARD T. KARSNER, M.D., LL.D.

RÉSUMÉ

ALL PARTS of the cardiovascular system may be affected in typhus, but there is variation from case to case in distribution and intensity of lesions. The topography and character of the lesions are strong indications of the presence of typhus, but that they are peculiar to this disease as compared with other infectious diseases cannot be positively asserted.

Capillary injury is conspicuous. In part, it is without morphologic change and is evidenced by hemorrhage as an indication of fragility or by edema or protein precipitate in the subcapsular space of renal glomeruli as an indication of increased

permeability. Swelling and proliferation of capillary endothelium are especially noteworthy in the dermis but occur elsewhere. Usually this change is accompanied by pericapillary cellular infiltration. Arterioles and venules may be similarly affected, though not so frequently as the capillaries.

Arteries of medium size occasionally exhibit cellular infiltration in intima, media, or adventitia, but only rarely is there necrosis of the wall. Veins are practically free of lesions, except that large veins may show cellular infiltration of the intima and thrombosis. Occasionally the intima of the aorta is the seat of cellular infiltration, rarely the adventitia. In all cases there is associated myocarditis of considerable degree. The media of the aorta is not similarly affected.

Interstitial myocarditis of slight, moderate, or profound degree is constant. Infiltration of mononuclear cells is frequent in the epicardial fat, and endocardium, and less frequent in the connective tissue of the epicardial surface.

CAIRO CASES

Gross Observations.—The subepicardial fat appeared gray and gelatinous in many patients. The myocardium was usually soft, flabby, brown, and opaque. In many of those dying after the 14th day of fever there were small opaque gray and grayish-yellow flecks and streaks in the myocardium that at times were confluent and resembled minute infarcts.

Microscopic Observations.—Capillaries: Hemorrhage from injured capillaries, swelling and proliferation of lining endothelium, perivascular infiltration by mononuclear cells, and thrombi were observed. Hemorrhage from capillaries showing no evidence of injury occurred in several instances, the best example being Case 16, where blood was present in the subcapsular space of a glomerulus and in neighboring tubules. In one instance blood was found in the periadrenal tissues without demonstrable morphologic lesions of the capillaries. Hemorrhage into pulmonary alveoli was frequently associated with pneumonia and was certainly related to capillary injury.

Swelling of capillary endothelium was apparently a general phenomenon and was occasionally accompanied by proliferation of cells as well.

Thrombi were frequent in capillaries. The material of which they were composed was finely granular or hyaline, only rarely containing a cell or two, and resembled platelets. It was always accompanied by a pericapillary cellular infiltrate. Occasionally there were strands of fibrin. Although not frequent, thrombosis was found in pulmonary capillaries in four patients. Thrombi were found most commonly in skin, tongue, myocardium, brain, spinal cord, kidneys, and periadrenal tissues, and less commonly in other tissues.

Cellular Infiltrations: The perivascular cellular infiltration included large mononuclear cells, lymphocytes, and plasma cells in variable proportions. For the most part it was around capillaries, but in certain instances it occurred around arterioles and small arteries and veins.

Pericapillary infiltration occurred in almost every tissue of the body. It was present in the skin and myocardium in most instances. In a few instances perivascular infiltrations showed tiny foci of necrosis of the exudate.

Diffuse infiltration of mononuclear leucocytes was encountered in many situations, usually, but by no means always, perivascularly. It was especially conspicuous in myocardium, tongue, meninges, and liver.

Nodules: The distinction between a focal infiltrate and a nodule was not sharp, but, when well outlined, as in the central nervous system, it is appropriate to speak of the lesion as a nodule. Similar nodules occurred in other organs, notably heart, lung, spleen, kidney, skin, muscle, and adrenal glands.

Arterioles and Venules: Perivascular infiltrates of mononuclear leucocytes occurred in six patients around arterioles rather than venules, but occasionally about both. In one instance there was necrosis of the exudate about a thrombosed arteriole deep in the corium, and in three instances mononuclear exudate appeared in the walls of arterioles.

Patchy necrosis of arterioles was accompanied by only little exudation, but there was a necrotizing exudative arteriolitis in three instances. In Case 22, the myocardium, epicardium, bone marrow, and capsule of adrenal glands showed fibrinois degeneration usually affecting only the adventitia, but in the bone marrow and just outside the adrenal capsule the media was involved as well.

As might be expected, exudative arteriolitis was usually accompanied by thrombosis, either occlusive or mural. Whereas thrombi in capillaries were granular or fibrinous, those in the arterioles contained cells which were more often mononuclear than polymorphonuclear. A few thrombi were unaccompanied by signs of inflammation.

Small veins were normal except for infiltration of mononuclear leucocytes in the intima of intrasplenic veins, a lesion common in other infectious diseases. Thrombi in venules, often with little or no perivascular infiltration, were observed in skin, esophagus, myocardium, adrenal gland, kidney, and testis.

Myocardium: The presence of mononuclear cells in the myocardium in extravascular positions justifies the view that an interstitial myocarditis was present. Lymphocytes, plasma cells, and large mononuclear cells, often with basophilic cytoplasm, occurred in varying proportions. Cytologically this might be called a subacute myocarditis, but, since it occurred in outspoken form as early as the sixth day, it may be termed acute. Often, but not invariably, the infiltration was perivascular. Focal obscurity of transverse striations was usual, even when autopsy was done only little more than an hour after death. Myocarditis was present in all but two patients and was especially marked in patients of Cases 3, 5, 7, 9, 16, and 23, dead respectively on the 11th, 12th, 11th, 14th, 9th, and 6th day. In other patients it was slight to moderate. Whether this means that in later cases the myocardial disease subsided or that death supervened early in patients with well-marked myocarditis cannot be asserted, but probably the latter is true. Dehydration was probably not the cause, as only patients of Cases 3 and 7 were said to be dehydrated and the patient of Case 9 was noted as well hydrated.

Epicardium: The epicardium was commonly involved. As in the myocardium, mononuclear cells were in part scattered diffusely in the fat and in part arranged around blood vessels. Vascular lesions also occurred.

Mural Endocardium: The mural endocardium was frequently acutely inflamed, but the valves were not attacked. The mural endocardium was focally infiltrated with considerable numbers of mononuclear cells, but there was no exudation of fibrin or of polymorphonuclear cells. The inflammation was usually greater in those patients with pronounced myocarditis. In one instance the base of the mitral valve at its attachment to the wall showed mucoid edema and swelling and fragmentation of elastica in addition to the cellular infiltration.

Aorta: Sections of aorta were included in material from 10 patients, 3 of which showed lesions provisionally attributed to typhus. There was a rich infiltration of mononuclear cells in the intima of two patients. This was also present in Case 26, together with perivascular infiltration in the adventitia associated with hemorrhage. In two patients there was syphilitic aortitis. In three patients it was normal. In the patients with no lesion the myocarditis was slight, but in two patients with definite intimal infiltration the myocarditis was profound. That intimal lesions are a feature of typhus is not certain, since similar intimal infiltrations occur in a variety of acute infectious diseases, but, in the latter, numerous polymorphonuclear leucocytes are usually present also. Until further studies of the aorta are possible the problem remains unsolved.

Arteries and Veins: Arteries and veins in the organs were affected in the same manner as the small vessels but in variable degree wherever observed in this material.

Healing: There were too few patients to justify any conclusion as to the process of healing, and only one with a fever lasting as long as 18 days (Case 21). The patient was 18 years old, and at autopsy the myocarditis was of moderate degree, but there was no infiltration in the epicardial fat and only slight infiltration in the endocardium. In Case 41, that of a patient aged 48 with 16 days of fever, the myocarditis was moderate, with cellular infiltration in epicardial fat and endocardium. In many instances of the same or shorter duration of fever, the myocardial involvement was no greater. Statistical evaluation was impossible, but there was no satisfactory evidence in favor of amelioration or healing of the myocarditis in patients who were febrile up to 18 days.

In a few instances there were fibrotic lesions. In one patient (Case 33), 35 years old, with fever of 12 days' duration, there was slight fibrous thickening of the intima of a coronary artery. In another patient (Case 41), who was 48 years old, with fever of 15 days' duration, there were foci of fibrosis in the myocardium. The same was true of the patient of Case 45, on which there were no clinical data. In this latter patient there was fibrosis with hyalinization in the endocardium. In the patient of Case 38, with no clinical data, there was fibrous thickening of the surface epicardium which appeared to represent an adhesion. To attribute these lesions to the healing of lesions of typhus was impossible. In the material available there were no indications of healing.

COMMENT ON LESIONS OF CARDIOVASCULAR SYSTEM

The changes in the cardiovascular system of patients who died during the Cairo epidemic of typhus were essentially the same as those described in other epidemics (Abrikosov, 1941; Allen and Spitz, 1945; Ceelen, 1919; Chiari, 1942; Durán, 1944; Golden, 1945; Randerath, 1943; Schopper, 1943; Wolbach, Todd, and Palfrey, 1922).

The fact that in epidemic typhus the initial lesion is chiefly in the endothelium of capillaries has been stressed by Wolbach (1948). Studies of the early stages of the disease in both man and laboratory animals have shown clearly that thrombi form in injured capillaries and propagate centripetally and that this process, which is favored by venous stasis, frequently results in necrosis. The involvement of capillaries, which are the most widely distributed of all the blood vessels, also

explains the diffuse character of the disease with lesions widely spread throughout the body (Wolbach, 1948). All authors are agreed that the large arteries of the heart are rarely affected and then only slightly. No reference to coronary occlusion attributable to epidemic typhus has been found, although Allen and Spitz (1945) observed a small nonocclusive thrombus in one coronary artery.

The nature of the cells in the perivascular accumulations has been debated. Our own observations are in agreement with recent evidence indicating that the cells are probably derived from both proliferation and exudation (Wolbach, 1948). This is shown by the localization of the lesions to the endothelium of blood vessels, the preponderance of macrophages over lymphocytes and plasma cells in the early stages, and by the presence of mitoses. Rickettsiae have been demonstrated in these cells (Wolbach, Todd, and Palfrey, 1922). The absence of significant numbers of polymorphonuclear leucocytes at any stage in the disease has been repeatedly noticed. Giant cells, which are thought to be binucleated plasma cells or macrophages, have been observed in an occasional patient (Allen and Spitz, 1945). Durán (1944) distinguished typhus nodules from Aschoff bodies by the presence of fibroblasts and eosinophiles and the eventual scar formation and anaphylactic nature of the latter. In our experience the perivascular cellular nodules were morphologically distinguishable from Aschoff bodies without difficulty.

The myocarditis is said to be most extensive in the papillary muscles and inner half of the adjacent ventricular wall (Wolbach, Todd, and Palfrey, 1922). In another study the left and right ventricles, the atria, and the interventricular septum were found to be equally attacked, although the involvement was usually focal (Allen and Spitz, 1945). Most authors have remarked on the apparent integrity of the myocardial fibers regardless of the severity of the lesion, except for fragmentation in the foci of cellular infiltrations. In the Cairo patients, however, atrophy, necrosis, and rupture of small numbers of myocardial fibers were observed. Vacuolization of myocardial fibers (Wolbach, Todd, and Palfrey, 1922), fatty degeneration of the myocardium (Chiari, 1942), and involvement of intramyocardial nerves (Allen and Spitz, 1945) have been described. Grossly the heart is usually normal in appearance, but it may be dilated, flabby, and friable (Chiari, 1942). Rickettsiae have not been demonstrated in myocardial fibers. Diphtheria may complicate typhus and produce secondary myocarditis (Schopper, 1943; Randerath, 1943).

Mural endocarditis of microscopic size has been frequently observed. Golden (1945) saw one patient with extensive fresh mural thrombi without evidence of underlying myocardial infarction, which he attributed to endocardial injury. He does not state whether the lesion was of macroscopic or microscopic dimensions.

No proved cases of valvular endocarditis due to epidemic typhus have been reported. Worms, Péquignot, and Held (1950) described a case of fatal endocarditis in a young man who had suffered from typhus five years before and at the time of his death had a positive Giroud-serum protection test for typhus. No other evidence of the cause of this lesion was given. The cases reported by Donzelot and Kaufman (1950) and redescribed by Donzelot, Kaufman, and Vernant (1950) are also not acceptable on the evidence presented.

It seems well established that large arteries may occasionally be the site of typhus lesions. Thrombosis of the superior mesenteric artery, with infarction of

the small intestine; thrombosis of the left internal carotid artery, with massive infarction of the left cerebral hemisphere; thrombosis of a main branch of the pulmonary artery; thrombosis of the splenic artery, with splenic infarction, and mural thrombosis of the aorta and common iliac arteries occurred in the Warsaw patients (Wolbach, Todd, and Palfrey, 1922). Microscopically the lesions took origin in the endothelium and consisted of swollen endothelial cells on or in the intima and were often accompanied by deposits of platelets and fibrin. These were regarded as specific typhus lesions and also as the starting point of the large thrombi.

Goodman (1935) suggested that typhus and thromboangiitis obliterans were causally related and described a skin test to help in making the diagnosis. Proof of this opinion is lacking.

Chiari (1942) has described "mesaortitis exanthematica" of the aorta and other large arteries that resembled syphilitic mesaortitis. Similar lesions were described many years ago by Ceelen (1919), but Wolbach, Todd, and Palfrey (1922) and Nicol (1919) failed to find them. It is not clear from the descriptions that concomitant syphilis was satisfactorily excluded by Chiari and Ceelen.

Acute thrombophlebitis of myocardial veins has been described by Allen and Spitz (1945) and by Chiari (1942). The latter author also observed similar lesions in the splenic veins. According to Allen and Spitz (1945), focal mononuclear phlebitis, usually with nonoccluding platelet thrombosis, is about as common as arteritis. Gouriou, Mondon, Marcon, and Lahillonne (1938) reported phlebitis of the lower extremity in an Abyssinian who had had epidemic typhus for 20 days. There were pain, swelling, edema, and increased heat in the affected leg, all of which subsided after a few days, except for the edema, which persisted for more than six months. Wolbach, Todd, and Palfrey (1922) observed mural thrombosis in the femoral and internal saphenous veins.

There is a paucity of information about the healing of vascular lesions. Abrikosov (1941) states that during convalescence the nodules in the brain and other organs disappear completely, the proliferated cells and cellular infiltrations being absorbed and the normal structure of the arterial wall restored. According to Randerath (1943), the typhus nodule disappears by the 31st day. These observations need confirmation. They suggest that the reason for the absence of significant residuals in many instances may be due to the fact that necrosis is not a constant or severe feature of the disease, whereas proliferation and exudations of inflammatory cells are common. Thus healing without significant scarring may be possible.

The clinical significance of the myocarditis is not well understood. At first it was thought to be the cause of the circulatory collapse, which is so common, but recently other explanations have been offered. Wolbach (1950) is of the opinion that diffuse involvement of sinusoidal endothelium, especially in spleen, liver and adrenal gland, is of importance in the production of such physiological disturbances as dehydration, loss of electrolytes from the circulation, fall in plasma volume and proteins, and peripheral circulatory collapse. German students of typhus during World War II thought that the cardiovascular manifestations of the disease were due to involvement of blood vessels in the vasomotor centers of the brain and that the effects of myocardial injury become apparent only late in the course of the disease (Laurentius, 1942; Randerath, 1943; Sturm, 1942). Most authors have been unable to show a good correlation between symptoms and myocardial lesions.

RESPIRATORY TRACT

ALVIN M. PAPPENHEIMER, M.D.

RÉSUMÉ

IT IS GENERALLY agreed that the pulmonary lesions found at autopsy in most cases of epidemic typhus are not specific and that only rarely are typical vascular lesions discovered. In the majority of cases there is bronchopneumonia, sometimes restricted to bronchioles, alveolar ducts, and adjacent parenchyma, sometimes extensive and confluent. The inflammatory exudate is characterized by a predominance of polymorphonuclear leucocytes and a variable amount of fibrin, edema, and hemorrhage. In a small number there is little or no alveolar exudate, but the septa are thickened and infiltrated with lymphocytes, plasma cells, large mononuclear cells, and a few polymorphonuclear cells. There are no hyaline membranes. To this lesion the term interstitial pneumonia is generally applied. In some instances the two types of pulmonary lesions occur concurrently in the same or different parts of the lung.

CAIRO CASES

It would seem unnecessary and repetitious to describe the lesions in individual cases. Of the 36 patients in this series in which lung sections were available, 23 had bronchopneumonia or confluent lobular consolidation, and 8 had a more or less pure type of interstitial pneumonia.

In one case there was massive diffuse consolidation with suppurative pleurisy, meningitis, and fibrinous pericarditis. With the Gram stain, pneumococci in great numbers were found in the exudate, and it was therefore regarded as one of Pneumococcus lobar pneumonia. Another was probably aspergillosis. There were granulomatous lesions with innumerable giant cells containing fragments of degenerating mycelium and areas of necrosis and suppuration. Finally, there were four cases in which there was no pneumonitis but only congestion, edema, emphysema, or focal atelectasis.

DEMONSTRATION OF RICKETTSIA-LIKE BODIES IN LUNG SECTIONS

Duplicate sections of the lungs from 33 patients were stained by Nyka's method [1] (1945) for rickettsiae and with buffered Giemsa and Gram stains for bacteria.

On the basis of histopathologic findings, the patients were divided into two groups. Group I comprised those patients in which the characteristic lesions of typhus were florid and widespread in brain, spinal cord, myocardium, skin, and other organs. Group II included those in which the lesions were less florid and restricted to a few organs. The Nyka-stained sections were searched for rickettsia-like bodies without knowledge of the group to which the patient had been assigned.

In 19 patients of Group I, rickettsia-like bodies were found in 13 (Table 2). In the 14 patients assigned to Group II, rickettsia-like bodies were seen in but 1 (Case 38). There seems thus to be a positive correlation between the presence of rickettsia-like bodies and the intensity and distribution of lesions in the body.

1. Fix in 10% formalin; stain 30 minutes to one hour in 1 : 10,000 aqueous methyl violet; differentiate in weak acetic acid (two drops of glacial acetic in 50 ml. H_2O); counterstain with 1 : 10,000 metanil yellow; wash, blot section, dehydrate in acetone, clear in xylene.

The intracytoplastic bodies discovered in the Nyka-stained preparations were identical in size and shape with Rickettsia prowazekii. Coccoid, diploid, and short bacillary forms were sharply stained purplish blue in contrast to the pure blue of nuclei. In most instances the bodies were present in great number and were unaccompanied by bacteria. In a few instances there was obviously a mixed infection. Contrary to expectation, the rickettsia-like bodies were found predominantly within the cytoplasm of polymorphonuclear leucocytes in the alveolar or bronchial exudate. Occasionally they were seen in large mononuclear cells either in alveoli or in thickened infiltrated septa. They were never found within bronchial epithelium, but, as was noted by Nyka (1945) in infected rat or mouse lungs, free extracellular forms, though not abundant, did occur.

In addition to the sharply stained and characteristic bodies described above, there were less clearly outlined coccoid bodies of somewhat larger size within polymorphonuclear leucocytes. These may have been degenerated or fused organisms altered in appearance by partial intracellular digestion.

Bacteria, when present, were easily distinguished from the rickettsia-like bodies by their large size and blue color.

Rickettsia-like bodies, chiefly within the cytoplasm of the polymorphonuclear leucocytes, were also well shown in preparations stained overnight with buffered Giemsa solution.

The absence of demonstrable bacteria in some instances and the finding of large numbers of rickettsia-like bodies might well be taken as evidence for the view that the pneumonia was due to rickettsiae rather than to bacteria. There are, however, certain considerations that impel reserve in accepting this conclusion. The Nyka method, although it stains rickettsiae admirably, is not specific, and to identify the bodies of rickettsiae solely on the bases of size and morphology is admittedly hazardous. This is true also of the Giemsa stain. With the Gram stain it is well known that ingested cocci in the course of intracellular digestion may lose their positive Gram staining, become reduced in size, assume pleomorphic forms, and, in short, closely simulate the appearance of rickettsiae. There is further the possibility that other small Gram-negative micro-organisms, such as Hemophilus influenzae, may be mistaken for rickettsiae.

Gram stains were made on duplicate sections of all lungs in which the Nyka stain had previously shown rickettsia-like bodies. With the exception of Cases 5 and 22, in which sparse numbers of Gram-positive cocci were found in the bronchial exudate, no Gram-positive organisms were present in the lesions. In a few instances large Gram-negative cocci (Neisseria catarrhalis?) were seen in the bronchial exudate along with the minute Gram-negative rickettsia-like forms.

The question may be raised at this point whether the failure to demonstrate Gram-positive cocci in the lesions may be due to overdecolorization with alcohol-acetone in the process of staining. Against this is the observation that, with the same technique, Gram-positive cocci were readily demonstrated on the surface of the pharynx or tonsils in a case in which sections of these tissues were available. The staining was further controlled by placing smears of Gram-positive streptococci and Gram-negative bacilli at the end of the slide and fixing them in 10% formalin.

Taken at their face value, our observations seem to show that rickettsia-like bodies may be present in great numbers within the leucocytes of the pneumonic exudate, without concomitant Gram-positive cocci. They cannot, therefore, be

interpreted as Gram-positive cocci which have become rickettsia-like in appearance because of intracellular digestion. Whether or not these bodies are really rickettsiae cannot be decided on purely morphologic grounds.

It is interesting to note that Stevenson and Balfour (1921), in a study of the respiratory lesion of typhus, expressed the same uncertainty. They state, "we are unable to differentiate the bodies found in the lung (Giemsa staining after fixation in Pick's solution) from the appearance regarded as rickettsiae," and again, "many of the polymorphs of the alveolar and bronchial exudate contain masses of organisms some obviously pneumococci, but others of a size and appearance suggesting the rickettsia type."

Azurophile intracellular granules have been described by Brug (1941) in smears and sections from nontyphus human lungs. Although Brug refers to the possibility of their being interpreted as rickettsiae, the colored drawings show no bacillary forms and the granules do not greatly resemble the bodies found in the inflammatory cells of the typhus lungs.

Ding (1944) was unable to demonstrate rickettsiae in sputum, nasal washings, or tracheal aspirates.

COMMENT

Support for the thesis that the R. prowazekii, unaided by secondary bacterial infection, is capable of causing bronchopneumonia may be found in animal experiments. By intranasal or intratracheal instillations of rickettsia-containing suspensions into mice, rats, or rabbits, it has been found possible to show extensive pneumonic consolidation and multiplication of rickettsiae in cells of the inflammatory exudate. Thus Castaneda (1939) by intranasal inoculation of suspensions of rickettsia-containing guinea pig tunica vaginalis or ground suspensions of infected lung tissue produced extensive hemorrhagic consolidation in lungs of mice, rats, and rabbits. Polymorphonuclear leucocytes were present in large numbers, and rickettsiae were found not only in bronchial epithelium but also in mononuclear and polymorphonuclear cells.

A more detailed histopathologic study employing various epidemic and murine strains administered by intranasal instillation into mice was made by Nyka (1945). Rickettsiae were present in enormous numbers both within cells and free in the exudate. Polymorphonuclear leucocytes were very numerous in the consolidated patches, and, while rickettsiae were most abundant in "alveolar" cells, they were seen in the cytoplasm of polymorphonuclear cells as well.

Similar observations have been reported by Ionesco-Mihaiesti and Ciuca (1943). After intranasal instillation of three to four drops of heavily infected mouse lung suspension, there was rapid mobilization of large numbers of polymorphonuclear cells in the capillaries and alveolar walls. These cells phagocytosed the rickettsiae which rapidly multiplied in the cytoplasm. Within 10 hours large mononuclear cells began to replace the polymorphonuclear cells and in turn were destroyed by multiplication of the rickettsiae. After about 40 to 48 hours the fixed endothelial cells of the arteries were invaded by rickettsiae, which multiplied profusely in the cytoplasm, often pushing the nucleus to one side and finally rupturing the cell wall. When these cells ruptured, they discharged their contents into the alveoli, which became filled with serosanguineous fluid rich in rickettsiae. In contrast to the findings of Castaneda, they were not taken up by the bronchial epithelium.

Avtsyn (1944) performed similar experiments and was able to elicit bronchopneumonia in mice by intranasal inoculation of rickettsiae-containing material. After 60 to 90 hours there was extensive consolidation.

Bablet and Girond (1944) showed that, after intratracheal injection of rickettsial infected suspension of rabbit lung, extensive bronchopneumonia develops in the early stages, of which the predominant inflammatory cell is the polymorphonuclear leucocyte. Rickettsiae were demonstrable in increasing number with progression of the lesions and were both intracellular and extracellular.

These experiments prove two things that bear upon the findings in this series of human cases: (*a*) Pulmonary lesions of bronchopneumonic type may readily be induced in laboratory animals by intranasal instillations of suspensions containing R. prowazekii, and secondary bacterial invaders play no part in their production; (*b*) rickettsiae are taken up and probably multiply within polymorphonuclear leucocytes, as well as macrophages.

Admitting that the route of infection in these experiments is by the upper respiratory passages, whereas in human patients the rickettsiae most probably reach the lungs through the blood stream, the fact remains that, once established, they are capable of eliciting a bronchopneumonic reaction without the intervention of streptococci or other bacteria and that they may be found in numbers within the polymorphonuclear cells that are a major component of the inflammatory exudate.

This is not to imply that secondary bacterial infection of the respiratory tract is not a frequent complication of typhus. Indeed, through the kindness of Prof. S. B. Wolbach, I had the opportunity to study sections prepared from 14 cases of epidemic typhus obtained in 1920 from the Warsaw epidemic and forming part of the material upon which the monograph of Wolbach, Todd, and Palfrey is based. Gram-positive cocci were present in all instances, and, although rickettsia-like bodies could also be found in six of the cases, they could not be distinguished from the bacteria which in the course of intracellular digestion became greatly reduced in size and pleomorphic, and lost more or less their capacity to retain the Gram stain. From material such as this, taken from patients who had received no antibiotics, it was reasonable to conclude that the terminal pneumonia must have been bacterial.

Aside from the types of pneumonitis described above, one may cite from the literature many examples of complicating bacterial infection of the respiratory tract: empyema (Killian and Obertries, 1943; Schopper, 1943); purulent perichondritis and edema of the glottis, sometimes fatal (Schopper, 1943). Meningitis, brain abscess, otitis media, mastoiditis, and septicemia have been observed to follow pulmonary infection (Schopper, 1943; Killian and Obertreis, 1943). Laryngeal, tracheal, and wound diphtheria may also occur in the course of typhus (Schopper, 1943; Randerath, 1943).

Such complicating bacterial infections did not occur in the Cairo material. Indeed, in the 33 cases in which lung sections were available, Gram-positive cocci were found in but 6 and in only 2 of the patients in which rickettsia-like bodies were present. Although it would be unjustified to draw final conclusions until the rickettsial nature of the organisms present in the pneumonic lesions has been established by biologic methods, the evidence provisionally supports the idea that R. prowazekii may evoke pulmonary lesions of the pneumonic type in the absence of complicating bacterial infection.

ALIMENTARY TRACT AND ASSOCIATED ORGANS

WILLIAM B. WARTMAN, M.D.

RÉSUMÉ

ALTHOUGH uncommon, lesions attributable to typhus have been described in all parts of the alimentary tract, with the possible exception of the colon and the salivary glands. Vascular and nodular lesions occur commonly in the tongue and palate and less commonly in esophagus, stomach, small intestine, liver, gall bladder, and pancreas. Swelling and proliferation of endothelial cells lining the sinusoids of the liver, acute hepatitis, and acinic dilatation in the pancreas are common. Complications include glossitis, sialadenitis, colitis, and various parasitic infections.

CAIRO CASES

Aside from petechiae, specific gross lesions were not discovered in the gastrointestinal tract at autopsy. Acute enteritis, necrotizing proctitis and colitis, and small ulcers in the colon were present but were thought to be unrelated to typhus. Some may have been due to infection with Schistosoma.

Tongue.—The glossal muscles were involved in 22 of the 27 cases in which material was available, showing typical nodules in 17 and vascular lesions in 19. The nodules consisted of swollen and proliferating sarcolemma, mononuclear cells, and plasma cells. Large arteries in the submucosa, often at the junction with the glossal muscles, were affected in 10 patients. In addition to nodules, there were diffuse accumulations of mononuclear cells, plasma cells, and lymphocytes in the subepithelial tissues in 20 patients. These were sometimes distributed about dilated capillaries in which the endothelium was distinctly swollen. Sections of tongue from Case 4 were stained for rickettsiae, but none were discovered.

About half of the patients showed moderate to marked parakeratosis and keratosis of the surface epithelium, which was crusted with granular debris, red and white blood cells, and bacteria. These changes did not differ from those seen in other febrile patients with a coated tongue.

Faucial Tonsil.—The tonsils were examined in 15 patients. Typical arteritis was present in one, and hyaline thrombi and nodules in another. About one-half showed latent acute or chronic tonsilitis of nonspecific type.

Palate.—Slides from six patients were examined. Changes in the pharyngeal mucosa were similar to those in the tongue. In one there was a large ulcer surrounded with mononuclear cells, but the blood vessels appeared normal. Hyaline thrombi were present in capillaries in five patients, and in one of them there was also arteritis. All showed moderate to marked acute inflammation.

Oropharynx.—The interstitial tissue in and about the mucous glands of the oropharynx was infiltrated with large mononuclear cells, plasma cells, and lymphocytes. In the material of one of the six patients examined there was slight hemorrhage in the periglandular tissue. In addition, the subepithelial tissue of the pharynx showed the same cellular infiltration and vascular lesions as were observed in the tongue. Perivascular infiltration of lymphocytes, large mononuclear cells, and plasma cells was prominent in some specimens.

Salivary Glands.—Slides were examined in 15 cases. In one a single arterial lesion was found in the substance of the gland; four showed slight chronic interstitial inflammation, and three showed slight dilatation of acini.

Esophagus.—Slides of the esophagus were available in six cases. In the muscularis of three of them there was a single artery showing necrosis and inflammation. In two of these there was sufficient exudate surrounding the involved artery to form a small nodule.

Stomach.—Slides were available from 26 patients, mostly from the pylorus. Of these, five showed mild to moderate vascular lesions in the muscularis as follows: typical arterial lesions, hyaline thrombi in the superficial capillaries of the mucosa, a subendothelial verruca in a capillary, bland cellular thrombi in the larger veins without inflammation of the wall. Nodules were present around affected vessels in three instances.

Small Intestine.—Slides from 25 patients were examined. The intestine showed changes similar to those in the stomach. A few hyaline thrombi in capillaries and nodular lesions were discovered in the mucularis in three patients. In addition, focal hemorrhage was noted in two patients, and in a few there was an increase of perigangliónic cells in the tunica muscularis. Cysts of schistosoma were found in one patient.

Large Intestine.—Slides from 18 patients were examined. Aside from nonspecific lesions such as schistosomiasis and necrotizing colitis, there were no significant changes in the colon. In a few there was an increase in the number of plasma cells in the mucosa.

Liver.—The liver weighed more than 2,000 gm. in 13 patients. The lobular pattern was preserved and the central zones were dark red and sometimes sunken.

There were sections of the liver for microscopic examination from 33 patients, of which 13 livers were normal and 4 were infected with Schistosoma. Vascular and nodular lesions were found in only two instances. In 15 there was marked diffuse swelling and proliferation of Kupffer cells, accompanied by exudation of fluid and an occasional neutrophile into the space of Disse between the parenchymal cells and the endothelial lining of the sinuses. The Kupffer cells showed conspicuous phagocytosis of red and white blood cells and infrequently a mitotic figure. Rarely a sinusoid contained a fibrin thrombus. Such reactive changes in the liver are, of course, not specific for typhus, since they are known to occur in other infectious diseases (Roessle, 1930). Small foci of necrosis of cord cells were present in eight instances. They were usually, but not always, in the midzone, and, since they contained considerable numbers of lymphocytes, plasma, and mononuclear cells, they resembled the nodules seen elsewhere in the body but could not be definitely established as such.

In most instances the cells about the central veins contained brown pigment that stained green azure eosinate, brown with ferrocyanide method, and deep orange red with oil red O.

The branches of the portal vein in two instances showed small subendothelial foci of hyaline material. The hepatic arterial branches in the portal spaces in most instances showed perivascular infiltration, and in two there were conspicuous arterial lesions. The bile ducts showed no deviation from normal.

Rickettsiae and inclusion bodies were searched for without success in tissues from patients of Cases 3, 4, and 7.

Gall Bladder.—No gross abnormalities of the gall bladder and extrahepatic bile ducts were noted.

Slides from 14 specimens were examined, and all but one was normal. In one a single large artery contained a small mural hyaline thrombus unaccompanied by inflammation.

Pancreas.—The pancreas and pancreatic ducts were normal on gross examination. Slides were available from 29 specimens. Characteristic arterial lesions showing hyaline thrombi or necrosis were found in three. In addition, there were other changes which were practically constant. These consisted of moderate to marked acinic dilatation, with retention of inspissated secretion. Occasionally, exfoliated and necrotic cells were found in the dilated acini. This lesion, which corresponded closely to that described by Baggenstoss (1948), was present in 23 instances. Occasionally it was accompanied by focal necrosis of acinic epithelium. In five patients there was also mild to moderate acute pancreatitis, with edema and polymorphonuclear leucocytes in the interstitial tissues. The inflammation did not involve the lumens of acini or the islets. In marked examples the acinic dilatation was diffuse, and in mild ones it was focal. In one instance there was moderate hyperplasia of islets.

COMMENT ON FINDINGS IN ALIMENTARY TRACT

The pronounced and almost constant involvement of glossal muscles, which was present in the Cairo patients, has not been described in other epidemics, probably because of failure to examine the tongue microscopically. This is another illustration of the frequent involvement of skeletal muscle in epidemic typhus and also confirms the statement of Wolbach, Todd, and Palfrey (1922) that "the involvement of large blood vessels in the skeletal muscle is second only to that in the skin and testis; although the lesions in muscle are not as striking as those in the central nervous system."

Numerous complications, mostly due to secondary bacterial infection, of the mouth and adjacent structures have been reported but were absent in the Cairo cases. These include abscesses in the floor of the mouth and ulceration of the arytenoid cartilage (Schopper, 1943) and purulent parotitis (Schopper, 1943; Randerath. 1943; Wolbach, Todd, and Palfrey, 1922), which is common in neglected patients and may be obviated by proper nursing care.

There is general agreement that the gastrointestinal tract is neither frequently nor markedly involved in epidemic typhus. Allen and Spitz (1945) encountered occasional small foci of thrombophlebitis in the stomach and colon, and Chiari (1942) saw central necrosis of the intestinal lymphoid follicles similar to that which occurs in the Malpighian follicles of the spleen.

Schistosomiasis was discovered in the colon in three of the Cairo patients and appeared to be clinically latent. Necrotizing colitis or proctitis was present in six patients, acute enteritis and esophagitis in one patient each, and ascariasis in one patient. Chiari (1942) described catarrhal colitis, especially of the cecum, and pointed out that clinically typhus may start with gastrointestinal symptoms.

The findings in the liver in the Cairo patients are much the same as those reported previously. Wolbach, Todd, and Palfrey (1922) noted the absence of lesions in large arteries and central veins. Occasionally slight to moderate phlebitis and arteritis of small vessels have been observed (Allen and Spitz, 1945). The central necrosis has been attributed to circulatory failure (Chiari, 1942). Schistosoma ova were discovered in the liver in four of the Cairo patients and diffuse

cirrhosis of Laennec's type in two. Almost no information is available concerning alterations of liver function. Wolbach (1948) believes that too little attention has been paid to the diffuse reaction of the sinusoidal endothelium of the liver and that this may explain in part the occurrence of hypoproteinemia and other functional disturbances. Chiari (1942) states that jaundice is not associated with the disease, but Schopper (1943) and Randerath (1943) observed jaundice in association with serous or parenchymatous hepatitis. However, the latter authors were unable to exclude the simultaneous existence of infectious hepatitis of viral or serum type.

Acinic dilatation and inspissation of pancreatic secretions, which was so common in the Cairo patients, has not been previously reported. In view of the fact that it is not uncommon in severe infections, it seems unlikely that it is a direct result of typhus (Baggenstoss, 1948).

URINARY TRACT

ALAN R. MORITZ, M.D., LL.D.

RÉSUMÉ

THE KIDNEYS share in the general distribution of blood vessel lesions and in the diffuse cellular infiltrations. The distribution of the lesions is spotty, and little of the parenchyma may be involved. The nodular perivascular aggregates of macrophages, plasma cells, and lymphocytes are most numerous in the cortex, and the diffuse infiltrates in the medulla are usually in relation to the vasa recta. In both instances blood vessels in or adjacent to the exudative zones appear to be the primary focus of injury. Vascular injury is both diverse and frequent, ranging from endothelial swelling to transmural necrosis. Hyaline thrombi are frequent in afferent glomerular arterioles and in some of the glomerular capillaries. Nonoccluding red and white thrombi are present in occasional arteries in most kidneys. Although the endothelium beneath such thrombi is characteristically destroyed or obscured, mural degenerative or inflammatory changes in relation to the thrombosis are not invariable. Hemorrhage may occur into the interstitial tissue and tubules, and blood may appear in the urine. Not infrequently this is found without demonstrated morphological vascular lesions. Hemorrhage also appears in the subcapsular space of the glomeruli without morphological lesion in the glomerular tufts. Degenerative changes in the nephrons with copius escape of protein-rich fluid and erythrocytes through the damaged glomerular tufts is seen with great frequency. Hyaline casts are constant in the lower portions of the nephrons. Small numbers of pigmented casts due to disintegration of erythrocytes are present in the loops of Henle and collecting tubules in about half the patients. These degenerative changes in the glomeruli and tubules are common to many other infectious diseases and must also be evaluated in the light of the degree of secondary bacterial infection, especially in the lungs.

CAIRO CASES

Kidneys.—The kidneys together weighed in excess of 350 gm. in 11 patients. The capsules stripped easily, and the outer surfaces were smooth and pale pink, and occasionally there were petechiae. The cut surfaces bulged, and the pattern was slightly obscured. The renal pelves, ureters, urinary bladder, and urethra showed only a few petechiae on gross examination.

Microscopic examination revealed changes in the parenchyma, the blood vessels, and the interstitial tissues of the kidneys.

Parenchymatous Changes: There was no example of outspoken acute glomerulonephritis, but in many instances either occasional or numerous glomeruli showed increased cellularity such as is commonly seen in acute infectious diseases. Some glomerular capillaries had narrowed lumens because of swelling of the endothelial cells. In a few there were focal lesions of single glomerular lobules consisting of hyaline thrombi and karyorrhexis. A variable amount of acidophilic precipitate was present in the glomular spaces. Some degree of degenerative change was observed in the epithelium of the proximal convoluted tubules in 35 of the 36 kidneys examined. In 17 instances focal or diffuse necrosis of the proximal convolutions was present. Frozen sections stained with oil red O showed fine fat droplets near the base of the cell. In nine instances the epithelium lining the collecting tubules showed hydropic degeneration, and the nuclei of occasional hydropic cells were pyknotic. There appeared to be no relation between hydropic changes in the collecting tubules and necrotizing changes in the proximal convolutions. In several instances swelling and granularity of the epithelium of loops of Henle and of distal convolutions were noted, but in no instance were the degenerative changes seen distal to the proximal convolutions of sufficient severity to warrant the diagnosis of lower nephron nephrosis.

Intraluminal granular precipitates or casts were seen in the nephrons of almost every kidney in the series. The distribution of protein precipitates in the various portions of the nephrons was as follows: glomerular spaces, 26 cases; proximal convolutions, 32 cases; loops of Henle, 34 cases; distal convolution, 22 cases; collecting tubules, 23 cases. In the majority of these the precipitate had consolidated sufficiently in the loops of Henle or in the collecting tubules to form hyaline or granular casts. In 22 instances extravasated erythrocytes were identified in some portion of the nephrons. In the upper portion of the nephrons the red blood cells were characteristically intact. In the lower portions of the nephrons most of the erythrocytes had disintegrated and were converted into granular pigmented casts. There appeared to be no association between the presence of casts in the lower portions of the nephrons and the occurrence of degenerative changes in the epithelium at that level.

Vascular Changes: Two types of vascular change were observed. In one the changes were mural and intraluminal and did not involve adjacent interstitial tissue to any significant degree. In the other the changes were both vascular and perivascular, and, because of the prominence of the perivascular element of the lesions, they will be described in relation to interstitial changes.

In the kidneys of 29 patients occasional thrombi were found in the lumens of arteries, arterioles, or veins. Rarely were they completely occlusive, and in no instance were they associated with infarction. Usually they gave the impression of being of recent origin. Although white thrombi were most frequent, some were red. Vessels of all sizes were affected and arteries more frequently than veins. Characteristically the endothelium where the thrombus was attached either had disappeared or consisted of disintegrating cells and nuclear fragments. In some instances the endothelium had started to grow over the edge of the free surface of the thrombus. Although degenerative and exudative changes were occasionally seen in the vascular

wall beneath the site of thrombus formation, these were exceptional, and in most instances the only mural abnormality that could be construed as antecedent to thrombus formation was swelling and degeneration of endothelial cells.

In the arterioles and the glomerular capillaries endothelial swelling was commonly seen, and the lumens of these vessels were often filled with an acellular coagulum of homogeneous acidophilic material.

Interstitial Changes: In 34 patients focal collections of exudative cells were found in the renal cortex, the medulla, or both. The outstanding features of these lesions were the relation of the cellular infiltrate to blood vessels and the character of the exudative cells.

In the cortex the infiltrates were characteristically nodular, were present at all levels but with a tendency to be most numerous in the juxtamedullary zone, and were most frequently situated around an afferent arteriole in proximity to a glomerulus. In some, a central blood vessel could not be recognized. A few nodules were found in the adventitia of large veins or arteries or in other situations where it was clear that the lesions were not in relation to an afferent glomerular arteriole. Individual nodules were one-fourth to twice the size of a glomerulus. The only fixed tissue structures that could be recognized as participating in their formation were the centrally placed blood vessels, which were usually arterioles. The lumens of the affected vessels ordinarily contained exudative cells and were encroached upon, but not occluded, by swollen intimal endothelial cells. Edema and degeneration of the subendothelial cement substance sometimes converted it into a thick-walled homogeneous acidophilic cylinder. The cells of the media in such instances were separated by the exudate.

In the medulla the exudative cells were more frequently disposed in ill-defined columns than in nodules. The endothelial cells lining the vasa recta which were adjacent to or incorporated within the region of exudation were swollen and hyperplastic, and the lumens of these vessels often appeared partially occluded by mononuclear cells. Although the collecting tubules and loops of Henle in such situations were often displaced, there was no convincing evidence that they were in any place disrupted or otherwise involved in the inflammatory reaction.

The individual cells comprising both the cortical nodules and the more diffuse medullary infiltrates varied from lymphocytes at one extreme to macrophages at the other, with plasma cells predominating. The nuclei of these cells were round or oval, with coarse, deeply staining aggregates of chromatin. Although the cytoplasm of these cells varied considerably in amount and in staining properties, it was characteristically abundant, homogeneous, and basophilic.

Search for Rickettsiae in Renal Lesions: Rickettsiae were searched for in specially stained sections of 17 kidneys. Clumps of minute coccobacillary rickettsia-like bodies were demonstrated in two kidneys, and single clumps not clearly identified as rickettsiae were seen in two kidneys.

Urinary Bladder.—Of 19 urinary bladders examined, specific arterial lesions were discovered in 3. Acute exudative or chronic "nodular" cystitis was present macroscopically in 11, and microscopic examination disclosed acute and chronic inflammatory changes associated with the presence of the ova of schistosoma in 9.

Ureters.—No lesions were found in microscopic sections from six patients.

Clinicopathological Correlation of State of Fluid Balance with Character, Severity, and Distribution of Renal Lesions.—The cases fell into four categories in respect to the available evidence relating to fluid balance.

1. Normally Hydrated: In 15 cases it could be inferred from the records that the patient was neither significantly dehydrated nor edematous at the time the observation was made.

2. Dehydrated: In 11 cases the patients were observed to be dehydrated at some time during their stay in the hospital.

3. Edema: In two cases edema was noted.

4. Indeterminate: In eight cases the data were insufficient to permit any conclusions in respect to fluid balance.

No feature of the pathological changes seen in the dehydrated group was peculiar to this group, or were there any changes in this group that did not occur with approximately equal frequency and intensity in the normally hydrated group. In 14 instances azotemia was recognized clinically. The changes observed in the kidneys of these persons did not differ significantly from those in whom azotemia was not recorded. No evidence of true glomerulonephritis was observed. Certain changes seen in the normally hydrated group were not recorded as being present in the dehydrated group. These included ischemia of the glomerular capillaries (four cases), an excessive number of mononuclear cells in the vasa recta (three cases), infiltration of the walls of veins by mononuclear cells (five cases), and interstitial cortical hemorrhage (five cases). The significance of these differences was not apparent. Since a person recorded as having a normal fluid balance in the hospital may have been recently hydrated after weeks of dehydration and a person recorded as being dehydrated in the hospital may have been so only during the last 24 hours of life, it was thought that the information available was not suitable for a clinicopathological correlation of this kind.

In most cases information regarding the duration of the disease and the duration of the patient's stay in the hospital was recorded. No relationship was discovered either between the state of dehydration and the duration of illness or between the duration of illness and the distribution or severity of the renal changes. Similarly there appeared to be no relationship between the pathological changes and either age or sex.

COMMENT ON LESIONS IN URINARY TRACT

There is general acceptance of the specific character of the vascular lesions and of the nodular and diffuse cellular infiltrations in the kidneys. Wolbach, Todd, and Palfrey (1922) observed that the lesions in the kidney do not show the proliferative reaction seen in the nodules in the skin and central nervous system but instead resemble the lesions in the myocardium in that only small vessels are affected and the cellular response is infiltrative rather than proliferative. These authors also found that hemorrhage into the interstitial tissues and tubules was almost constant in the presence of acute lesions and that those kidneys that did not show them were from patients dying between the 7th and 18th day of typhus.

The nature of the glomerular and tubular changes, however, is not clear. Several authors have stated that acute diffuse hemorrhagic glomerulonephritis may be a frequent complication of epidemic typhus. Randerath (1943) reported it in 75%

of cases occurring over a 16-month period in the civilian population of an occupied Soviet territory. He remarked that most of the patients with glomerulonephritis were seen during the winter. Herzog (1935) claimed the incidence of glomerulonephritis as 65% of fatal cases in an epidemic in Chile. Another 23% were said to have interstitial nephritis and only 10% to have no kidney lesions. Caffarena (1937) found acute diffuse glomerulonephritis in 66% of his cases. Wetzel (1940) described a fatal case of typhus in which there was acute glomerulonephritis and expressed the opinion that renal injury had not been sufficiently emphasized. Dawydowskie (1923) noted focal destructive glomerulonephritis in one-third of his cases. Unfortunately the pathological descriptions and illustrations in these papers do not permit objective evaluation, and it is not clear that the authors differentiated beween the lesions of classical acute diffuse hemorrhagic glomerulonephritis and those of epidemic typhus. Allen and Spitz (1945) reported the intracapillary form of acute diffuse glomerulonephritis in 18 cases, acute focal glomerulitis in 3, and normal glomeruli in 2 of 23 cases. They suggested that the glomerular alteration might be regarded "as a remote effect of the rickettsiae—either hyperergic or toxic—in much the same sense that acute diffuse glomerulonephritis following scarlet fever is attributed to the distant effects of Streptococcus hemolyticus." Similar views had been expressed by Julliard and Henaff (1939). Allen and Spitz (1945) were also of the belief that the renal lesions were reversible in patients recovering from the disease.

Since the material which Allen and Spitz (1945) studied was obtained from typhus cases of the Cairo epidemic, we have taken the opportunity to review it. As a result of this, as well as of a careful study of Allen and Spitz's descriptions and illustrations and of a study of additional cases, we are of the opinion that the Cairo patients did not have acute diffuse glomerulonephritis in the usually accepted use of that term. What they did have was slight to moderate swelling of glomerular capillary endothelium similar to that observed in many severe, acute infectious diseases. It seems reasonable to believe that such alterations in the glomerulus are reversible, but proof of this opinion is lacking. We could discover no objective evidence to support the suggestion that they were due to toxins or hypersensitivity.

Other authors also have failed to find evidences of acute diffuse glomerulonephritis. Chiari (1942) never saw altered glomeruli in early cases and Schopper (1943) found that in most of his cases there was a variable amount of hematuria but without anatomical signs of glomerulonephritis. He also pointed out that the presence of red blood cells and swollen and proliferating endothelium in Bowman's capsule might present a histological picture resembling that of acute glomerulonephritis and that such changes were most marked when septic complications were at their height. This same observation was made many years ago by Wolbach, Todd, and Palfrey (1922), who also commented that proliferation of intracapillary glomerular epithelium is not to be regarded as characteristic of typhus since it may occur in other infectious diseases, notably influenza.

There are two recent reports (Allen and Spitz, 1945; Golden, 1945) of pigmented casts in the distal convoluted tubules. On the basis of tinctorial reactions these have been thought to be hemoglobin casts, but, in view of the unproved specificity of so-called hemoglobin stains, this conclusion cannot be accepted as proved. Also recent studies by Wheelock and Teloh (1952) show a high incidence of

morphological and tinctorially similar casts in routine autopsies. The Cairo material does not lend support to the belief that a lower nephron nephrosis occurs in epidemic typhus.

The changes in the other portions of the urinary tract are not remarkable. No description has been found of specific lesions occurring in the male or female urethra or in the penis.

FEMALE GENITAL TRACT

ROBERT A. MOORE, M.D., Ph.D.

THE FEMALE genital tract is usually spared or only slightly affected in epidemic typhus. In the Cairo patients the organs were normal grossly, but on microscopic examination minimal and inconspicuous lesions were discovered in the vagina (two examined), uterine cervix (one examined), fundus of the uterus (five examined), and ovary (six examined). The lesions observed were slight lymphocytic infiltration of the vaginal wall in two patients, a specific arteriolar lesion in the vaginal wall of one patient, scattered lymphocytic infiltration of the cervical and fundal myometrium in one instance each, dilatation of endometrial sinusoids in one patient, hyaline thrombi in the superficial sinusoids of the endometrium with slight hemorrhage in one patient, and a few specific arteriolar lesions in one ovary.

Wolbach, Todd, and Palfrey (1922) examined the following organs microscopically with negative results: breast (6), uterus (6), ovaries (16), and Fallopian tubes (5). On the basis of experiments on two pregnant guinea pigs, Combiesco, Popesco, Stammatesco, and Angelesco (1934) claim that the placenta is pervious to R. prowasekii.

MALE GENITAL TRACT

ROBERT A. MOORE, M.D., Ph.D.

RÉSUMÉ

THE TESTIS, epididymis, and skin of the scrotum are commonly affected in epidemic typhus, exhibiting characteristic vascular lesions, nodular and diffuse cellular infiltrations, and necrosis. The prostate, seminal vesicles, and spermatic cord are attacked less severely and much less frequently. Degenerative changes, which are often marked, and atrophy of the epithelial cells of the testicular tubules, together with edema and fibrosis of the interstitial tissues, are almost constant findings. Aspermatogenesis and increase in Leydig's cells may be seen. Except for the skin of the scrotum, which may show grossly visible lesions like those elsewhere in the skin, the changes are detected only on microscopic examination.

CAIRO CASES

Testis.—Testes of 16 persons were examined. In all testes from those past puberty there was some suppression of secondary spermatogenesis. This was characterized by the presence of small seminiferous tubules; thickening of the basement membrane; decrease of spermatids and spermatazoa in the tubules, with karyorrhexis of those remaining; rare multinucleated cells in the lumen, and almost complete absence of mitoses in spermatogonia. In other words, these changes were typical of those observed in most of the acute infectious diseases and were not specific.

Specific lesions were of two types, typical changes in arterioles and focal interstitial orchitis. The lesions of arterioles, present in 11 patients, were like those in other organs.

Focal interstitial orchitis was usually of slight to moderate grade and consisted of infiltration of large mononuclear cells, plasma cells, and lymphocytes about dilated capillaries. The inflammation did not involve seminiferous tubules, and in no instance was there necrosis of the focus. This type of change was present in 12 of the 16 testes examined.

Independent of the degree of vascular change and the interstitial orchitis, there was an interstitial edema in some. There were no alterations in the interstitial cells.

Prostate.—Twenty-one prostate glands were examined. The architectural pattern and structure of the epithelial and stromal cells were normal for the age of the patient, except for moderate focal epithelial atrophy and slight degenerative changes in the smooth muscle cells of the stroma which is consistent with an acute infectious disease.

In the richly vascular stroma about the distal part of the intraprostatic urethra and in the tissues anterior to the urethra there was in most instances slight to moderate focal infiltration with lymphocytes, plasma cells, and large mononuclear cells most prominently observed about medium-sized sinusoidal vascular spaces but rarely about capillaries. In one instance there was a thrombus in the sinusoid. Because this type of infiltration is rarely seen in the prostate in other diseases and inasmuch as the types of cell were the same as those present in other tissues in typhus, it is believed to be a specific lesion.

Within the body of the prostate there were instances of lymphocytic infiltration, but these did not differ from the usual picture of so-called "chronic prostatitis," and hence no significance was attached to them. In one instance the focal cellular infiltration in the stroma was of specific type. In two instances there was necrotizing purulent urethritis, with spread into the prostate.

In four instances there were specific arteriolar lesions with necrosis, thrombosis, and cellular infiltration.

Bland thrombi in the periprostatic veins were of the type frequently seen. They were prominent in one patient.

Seminal Vesicles.—Seminal vesicles of nine patients were examined. A single vascular lesion was found in one. Except for focal infiltration with mononuclear cells in one instance, which was of doubtful specific significance, there were no other pertinent changes. In four vesicles there was fibrosis of the wall associated with calcified ova of schistosoma.

Spermatic Cord.—Of the five spermatic cords examined, lesions included slight cellular infiltration in four, bland thrombi in the venous plexus in two, and specific arterial lesions in one. The cremasteric muscle showed cellular infiltration in one instance.

COMMENT ON LESIONS IN MALE GENITAL TRACT

The findings in the Cairo cases are identical with those reported in other epidemics of typhus. In addition, slight hyaline thickening of the basement membrane of the testicular tubules (Wolbach, Todd, and Palfrey, 1922) and involvement of the tunica albuginea (Allen and Spitz, 1945; Golden, 1945) have been observed. The results of this and of other studies (Allen and Spitz, 1945; Durán, 1944; Golden, 1945; and Wolbach, Todd, and Palfrey, 1922) clearly indicate that testicular involvement occurs in epidemic typhus about as frequently as in Rocky Mountain spotted

fever and other rickettsial diseases and that its presence is not of help in differential diagnosis. Wolbach, Todd, and Palfrey (1922) were able to demonstrate rickettsiae in the endothelium of blood vessels in Giemsa-stained sections of the testis or epididymis in five instances.

HEMATOPOIETIC TISSUES

EDWARD B. SMITH, M.D.

RÉSUMÉ

SPECIFIC lesions are rarely found in the tissues of the hematopoietic system. Splenomegaly is often present, and microscopically the follicles are small and inconspicuous, and there is pronounced erythrophagocytosis. Infiltrations of mononuclear cells may be seen in the adventitia of the intratrabecular arteries and beneath the intima of the intratrabecular veins. The cells lining the sinusoids of lymph nodes and the bone marrow may become moderately hyperplastic. Usually there is a neutrophilic leucocytosis with a shift to the left, but the hemoglobin and the number and appearance of the red blood cells are normal. It seems likely that the lesions in the hematopoietic tissues are, at least in part, dependent upon the presence of secondary bacterial infection.

CAIRO CASES

At autopsy the spleen weighed more than 250 gm. in 22 patients. The pulp was moist, dark red, frequently soft, and occasionally almost fluid. The follicles usually were not recognizable. A thin fibrinous exudate covered the capsule in two instances.

In a few patients the lymph nodes were slightly enlarged, gray, and soft.

The bone marrow of ribs, sternum, vertebrae, and skull was dark red.

Spleen.—The histopathologic pattern bore out the gross findings of an enlarged soft dark red spleen. The red pulp was greatly increased owing to excessive numbers of erythrocytes between sinusoids. In the congested pulp, lymphocytes were decreased in number, but there were numerous plasma cells, some of which were binucleate. Of note was the almost constant absence of neutrophilic and eosinophilic granulocytes. A few megakaryocytes were regularly seen in the pulp and sinusoids. Slight fibrosis, found in one-third of the patients, was regarded as an incidental finding.

The sinusoids were inconspicuous and lined by slightly enlarged endothelial cells. An occasional large mononuclear cell of the endothelium or a reticulum cell in the red pulp contained erythrocytes. Erythrophagocytosis was present in moderate amount in virtually all spleens. This was most easily demonstrated by oil-immersion microscopy after location of a large mononuclear cell under "high dry." Without oil immersion the phagocytes merely presented a diffuse color of hemoglobin due to disintegration of overlapping erythrocytes. An occasional spleen contained hemosiderin particles.

A recently formed thrombus was present in a sinusoid of three patients. Another patient presented a small recent infarct. Otherwise, no evidence of vascular obstruction in the spleen was discovered.

The follicles were small and rarely showed secondary nodules. Neither spleen nor lymph nodes displayed reactive centers. An occasional enlarged reticulum cell appeared in a follicle merely as a part of generalized activity of the reticuloendothelial

system. In one-third of the spleens a few follicles contained deposits of coarse, hyaline, fibrin-like material in which were mononuclear cells and an occasional neutrophilic granulocyte. These deposits were regarded as nonspecific and were most numerous in cases of 10 days' or more duration. Hyalinization of the central arteriole was present in about two-thirds of the 30 spleens examined.

Intratrabecular blood vessels were usually normal, but in one-third of the spleens there was slight mobilization of mononuclear cells in the adventitia and beneath the intima of intratrabecular veins. The trabeculae and capsule were devoid of inflammatory cells.

Lymph Nodes.—Histologically the lymph nodes showed an inconstant hyperplasia which, when present, affected chiefly the lining cells of the sinusoids. In a reaction of moderate degree the sinusoids were conspicuous as anastomosing broad channels in which fixed enlarged lightly staining littoral cells and fixed macrophages predominated. However, in some instances there were numerous rounded monocytes lying free in the sinusoids. These free cells exhibited erythrophagocytosis more frequently than the fixed cells, and occasionally leucophagocytosis was observed. The medullary cords and follicles were virtually devoid of reaction. In fact, the latter were usually without secondary follicles or nodules and were strikingly few and small. The capsule was usually intact.

Bone Marrow.—Sections of bone marrow were available from 31 patients and displayed hyperplasia in which most of the fatty marrow (of vertebrae) was replaced. Only four specimens showed less than 75% of the marrow space occupied by cells; half of the marrow sections were more than 85% cellular. The increase involved all blood-forming elements with least change in erythropoietic cells. The myelocytic series and the megakaryocytes were moderately hyperplastic. The latter were approximately doubled in number per cubic millimeter of bone marrow as calculated from the number found in 100 high-power fields in each instance. There was a proliferation of endothelial phagocytes in capillaries and among the hematopoietic cells. The cytoplasm of these cells was sometimes so engorged with erythrocytes that it might easily be mistaken for the cross section of a congested small capillary. Erythrophagocytosis was ordinarily not excessive, even though 27 of the 31 specimens available showed the phenomenon. The relationship of leucophagocytosis to the absence of leucocytosis was unexplained. A few specimens showed occasional small foci of necrosis or collections of mononuclear cells without, however, demonstrable relation to blood vessels.

The age of the patient did not alter the pattern of reaction of the bone marrow, and it was not possible to determine the duration of the illness by the changes noted. The characteristic vasculitis of typhus was not usually detected in the marrow.

COMMENT ON LESIONS OF HEMATOPOIETIC TISSUES

Wolbach (1948) has shown that the histology of the spleen varies with the duration of the disease and the presence or absence of bacterial pneumonia or other infectious complication. In the early periods of the disease he describes marked engorgement with blood, lymphoid depletion of the pulp, inactive follicles, and a great increase of macrophages in the sinuses and reticular cords. Erythrophagocytosis may be considerable (Wolbach, Todd, and Palfrey, 1922; Allen and Spitz,

1945; Schopper, 1943) but is usually less marked than in typhoid fever (Wolbach, Todd, and Palfrey, 1922). Chiari (1942) was unable to demonstrate erythrophagocytosis in his cases. Late in the course of the disease hemosiderin may be abundant (Wolbach, Todd, and Palfrey, 1922; Chiari, 1942), and many plasma cells may be found at the periphery of the follicles and in the pulp. Considerable numbers of polymorphonuclear leucocytes may appear later or earlier if extensive pneumonia is present. Myeloid transformation, as reported by Nicol (1919), has not been observed by recent workers. Terminal hemorrhagic foci and zonal necrosis have been reported (Golden, 1945). Destruction of nuclei with formation of Fleming's bodies in the Malpighian follicles has been described (Chiari, 1942). Rickettsiae are said to have been demonstrated in one case by Allen and Spitz (1945), but others, including ourselves, have failed (Wolbach, Todd, and Palfrey, 1922). Experimentally in both rabbits and guinea pigs, when the spleen was removed and the reticuloendothelial system blocked with India ink, high fever and severe lesions in brain and viscera followed infection with R. prowasekii. Control animals showed only inapparent infection (Jelin, Linetzkaja, and Grossmann, 1934).

There is general agreement that the lymph nodes undergo some degree of hyperplasia with an increase in the number of macrophages and that the bone marrow shows similar hyperplasia and, in addition, phagocytosis of both red and white blood cells by macrophages. The hyperplasia involves all elements of the marrow and cannot be distinguished from that which is present in secondary anemia (Wolbach, Todd, and Palfrey, 1922). In smears of marrow obtained by sternal puncture, Benhamou (1942) saw a great increase in the number of endothelial cells and a still greater increase in plasmocytes. There were 10 to 20% of lymphocytes instead of the normal 3 to 5% and 2 to 3% megakaryocytes in place of the normal 1%. The percentages of myelocytes, polymorphonuclear leucocytes, and erythroblasts were correspondingly reduced.

The alterations of the cellular constituents of the blood are summarized by Yeomans (1948): "The white blood cell count is frequently reduced during the first week of typhus. The differential count may show a relative lymphocytosis. The white count may become elevated during the second week, particularly at the time of efflorescence of the rash. At all times during the febrile illness the eosinophiles are either absent or much reduced in number. A reduction in the red cell count is frequently seen during the clinical coure of the disease. Counts of 3-3.5 million cells per cubic millimeter are not uncommon. The anemia appears to be of the normochromic type. With no special therapy the red cell count returns to normal values in early convalescence." Similar observations have been made by others (Wolbach, Todd, and Palfrey, 1922; Danielopolu and Craciun, 1939; Milhaljevič and Radičev, 1942; Lampert, 1943; Allen and Spitz, 1945). Wolbach, Todd, and Palfrey (1922) were unable to demonstrate rickettsiae in either thick or thin blood smears taken from the finger blood of 167 patients at all periods of the disease. Epstein, Turewitsch, and Exemplarskaja (1934), by the use of special differential staining methods, claimed to have demonstrated rickettsiae in monocytes in the blood of typhus patients. Allen and Spitz (1945) described sickled red blood cells in the renal veins of tissues from Egyptian patients who died from epidemic typhus. There were no perifollicular pools of blood in the spleen.

ENDOCRINE GLANDS

ROBERT A. MOORE, M.D., Ph.D.

RÉSUMÉ

WITH THE possible exception of the adrenals, the endocrine glands are neither regularly nor severely attacked in epidemic typhus. Occasional vascular lesions and nodular or diffuse infiltrations of mononuclear cells have been described in the nerve portion of the pituitary and in the thyroid, parathyroids, and adrenals. The thymus and pancreatic islets are apparently spared. For changes that occur in the testis, see the section on the genital system.

Involvement of the adrenal cortex is fairly common and includes occasional typical blood vascular lesions and foci of mononuclear cell infiltration. In addition, there may be foci of lipid depletion, of lymphoid and plasma cells, and of degeneration of the cells of the fascicular zone. These changes may be very slight or absent and, except for the specific lesions, are such as may occur in any infectious disease.

CAIRO CASES

Adrenal Gland.—The adrenal glands of 31 patients were examined. At autopsy the outer cortical zone contained much less lipid than normal and was gray and translucent. This finding was not uniform, since small islands of yellow lipid-containing cells persisted here and there in the cortex.

Microscopic examination of the cortical cells, especially in the glomerular zone, which was increased in thickness, showed changes usually observed in acute infectious diseases, namely, depletion of fat, and necrosis of some cells, leading to the formation of pseudoglands. Frozen sections stained with oil red O, despite storage of the blocks in alcohol, showed fine droplets of fat in greater or less abundance in the cells of the reticular zone and to a less extent in the fascicular zone.

Focal infiltration with lymphocytes, large mononuclear cells, and plasma cells was observed about the central vein and its branches, in the medulla, and in the inner part of the cortex. In the reticular zone there were nodular collections of mononuclear cells. Rarely the endothelial cells of the sinusoids were slightly swollen. In some instances the cellular exudate in the cortex showed signs of early necrosis.

The chromaffin cells of the medulla showed no visible change.

Irregularly throughout the capsule and periadrenal tissue there was diffuse and perivascular infiltration with large mononuclear cells, lymphocytes, and plasma cells. Occasional arterioles showed typical lesions of typhus, including necrotizing vasculitis and fibrinoid change.

Pituitary Gland.—In the six specimens available, no alterations of the anterior lobe were demonstrable. In three specimens the posterior lobe showed nodules comparable to those in the brain. In addition, in one instance there were swelling and proliferation of capillary endothelium, hyaline thrombi in capillaries, and slight perivascular infiltration with lymphocytes.

Thyroid Gland.—Twenty specimens were examined. Perivascular lymphocytic infiltration was occasionally present in the interstitial tissues and capsule. In one instance there was infiltration of mononuclear cells in the wall of a small artery.

Parathyroid Gland.—Two glands from different patients were examined, and there was one perivascular focus of lymphocytes in the interlobular septum.

Thymus.—The thymus in the eight specimens available showed advanced involution. Hassall's corpuscles were frequently necrotic and cystic. No cellular infiltration and no lesions of blood vessels were observed. The arteries in the fat about the thymus in two patients, aged 28 and 35 years, showed calcification of the elastic fibers.

COMMENT ON LESIONS OF ENDOCRINE GLANDS

Some authors have described more extensive necrosis of the adrenocortical cells (Golden, 1945) and more constant tubular degeneration in the fascicular cords (Allen and Spitz, 1945) than were observed in either the Cairo cases or other reported series. However, these authors noted that similar lesions have been described in a variety of acute infections (Rich, 1944) and did not attribute them specifically to typhus. Depression of eosinophiles during the febrile period of typhus has been reported (Yeomans, 1948). The difficulty of interpreting the morphological changes in the adrenal gland is well expressed by Wolbach (1948): "There is a temptation to associate these spotty and relatively minor cortical lesions with the low blood pressure or 'circulatory collapse,' so characteristic of severe cases of rickettsial disease, but no correlation has been made showing relationship between degree of adrenal damage and degree of circulatory collapse, nor has any evidence of adrenal inadequacy been shown by means of chemical or physiological studies, or by the substitution therapy suggested on the basis of adrenal cortical insufficiency. . . . Cortical lesions of the adrenal may be very slight or absent. When extensive lesions are encountered in the post-mortem study of rickettsial diseases, they should be appraised in the light of secondary bacterial infection of the lungs."

The observation made in the Cairo cases that the anterior lobe of the pituitary gland is spared, whereas typical Fraenkel nodules may develop in the posterior lobe, was also made by Wolbach, Todd, and Palfrey (1922). They found the nodules around capillaries and observed no participation on the part of neuroglial cells. They also saw perivascular accumulations of macrophages, lymphocytes, and plasma cells in the substance of the posterior lobe and in the investing dura.

MUSCULOSKELETAL SYSTEM

ROBERT A. MOORE, M.D., Ph.D.

RÉSUMÉ

THE STRIATED muscles are almost constantly affected in epidemic typhus, especially in the heart, tongue, and diaphragm. Not only are typical vascular lesions common but also the involvement of large blood vessels in skeletal muscle is second only to that in skin and testis (Wolbach, Todd, and Palfrey, 1922). Interstitial myositis and both nodular and diffuse accumulations of mononuclear cells are frequent. In contrast, involvement of smooth muscle is infrequent and characteristically slight. No information is available concerning possible involvement of tendons, joints, bones, and cartilages. Alterations of the bone marrow have been described in the section on the hematopoietic system.

CAIRO CASES

No gross lesions were found at autopsy in the musculoskeletal system, with the exception of symmetrical ischemic necrosis (dry gangrene) of the feet in one instance.

Skeletal Muscle.—It is significant that microscopically there were lesions in striated muscle regardless of the site of origin. Thus, either specific vascular lesions or focal interstitial myositis or both were present in striated muscle from the tongue, the heart, the neck about the thyroid, the diaphragm, the anterior part of the prostate, and about the pharynx.

The lesions in arterioles and capillaries were the same as in other parts of the body. Interstitial myositis appeared in two forms. One type resembled the usual focal interstitial myositis, with edema, increase of sarcolemmal nuclei, capillary dilatation, and infiltration of large mononuclear cells, plasma cells, and lymphocytes. The second type consisted of focal leucostasis in the capillaries, with necrosis of the fibers supplied by them.

In general, the two types of lesion were more common in the tongue than elsewhere.

Smooth Muscle.—Vascular lesions and nodules were discovered in the smooth muscle of the esophagus, stomach, and small intestine. However, they were small in size and number and seen in only a few cases. No lesions were discovered in the smooth muscle of bronchi, colon, uterus, Fallopian tubes, prostate, urinary bladder, ureters, bile ducts, or gall bladder.

Bone.—In one instance a section of bone of unknown site was available for microscopic examination and showed a single small arterial lesion in the periosteum.

LESIONS IN MUSCULOSKELETAL SYSTEM

Brittleness of the skeletal muscles and a dry cloudy appearance in the fresh state have been described (Aschoff, 1915; Chiari, 1943), but others have not observed it (Wolbach, Todd, and Palfrey, 1922). Various degenerative changes in the muscle fibers have been reported, including Zenker's waxy degeneration (Aschoff, 1915; Wolbach, Todd, and Palfrey, 1922; Chiari, 1943; Schopper, 1943). Hemorrhage and necrosis occur rarely (Chiari, 1943). In late cases regeneration of sarcolemma may be observed (Wolbach, Todd, and Palfrey, 1922). Specific involvement of the following muscles has been reported: biceps and quadriceps extensor group (Wolbach, Todd, and Palfrey, 1922); rectus abdominalis (Wolbach, Todd, and Palfrey, 1922; Schopper, 1943); tongue (Allen and Spitz, 1945; Report of National Research Council, 1953); diaphragm (Allen and Spitz, 1945; Report of National Research Council, 1953); muscles of the lower extremities (Schopper, 1943), and muscles about the pharynx, thyroid, and anterior portion of the prostate (Report of National Research Council, 1953). Doubtless, if other muscles were examined microscopically, they too would be shown to be attacked. The apparent sparing of tendons, joints, bones, and cartilages may also be explained by failure to examine these structures rather than to some special feature of the disease.

Massive necrosis of the skin and subcutaneous tissues or of a whole or a part of an extremity is frequent in epidemic typhus, but opinion differs as to its explanation. Wolbach, Todd, and Palfrey (1922) studied the lesion in six of their patients and came to the conclusion that the large areas of necrosis of the skin (gangrene), whether or not symmetrically distributed, were not accompanied by thrombosis of large vessels. Microscopic study showed thrombosis of capillaries, small arteries, and veins, beginning in the corium and extending centripetally, which they believed was the probable cause of the necrosis. Stasis due to pressure was thought to favor

the localization of the endangiitis of typhus. Symmetrical gangrene was attributed to nerve lesions. Gangrene has also been attributed to spasm, exposure to cold, nodules in the vasomotor centers of the brain (Sylla, 1942), and nodules or thrombi in both small and large arteries (Killian and Obertreis, 1943). Despite its serious nature, this complication of epidemic typhus has been little studied, the only recent investigation being that of Schopper (1942), who examined material obtained both at autopsy and from amputated legs. The cases occurred in the civilian and military population of Soviet territory occupied by the Germans during World War II. Unfortunately, many of the patients also suffered from the effects of exposure to extreme cold and had frostbitten or frozen extremities, and Schopper could not always distinguish between lesions due to cold and to typhus. On the basis of this material he concluded that the primary incident responsible for the development of gangrene in typhus was failure of the circulation due to involvement of the vasomotor centers in the brain. Clinically this was manifested three to four days before death, when the lower extremities became cold, cyanotic, and more or less pulseless, although the tissues remained viable. Pathological examination of such extremities revealed greatly narrowed arteries, presumably due to spasm, which, however, were free of thrombi or emboli. If an additional injury had occurred during life, such as stenosing angiitis or pressure decubitus in typhus or thrombosis of small arteries and veins in cold injury, then massive necrosis or gangrene developed. Thus gangrene and massive necrosis, according to Schopper, are due to a combination of circulatory failure and local vascular injury involving principally small vessels. However, it must also be remembered that thrombosis of large arteries with massive infarction occurs (Wolbach, Todd, and Palfrey, 1922; Sylla, 1942). Embolism can usually be excluded, although Schopper (1943) described a rare case in which gangrene of a lower extremity was due to emboli arising from a large mural thrombus in the left ventricle, which he attributed to typhus. There was no medial calcification of the leg arteries in this case.

(To Be Concluded)

News and Comment

American Academy of Forensic Sciences.—The American Academy of Forensic Sciences will hold its Sixth Annual Meeting, Feb. 25, 26, and 27, 1954, at the Drake Hotel, Chicago. Titles of all papers to be read must be submitted to Dr. Milton Helpern, Program Chairman, 106 E. 85th Street, New York 28, Nov. 1, 1953.

Dr. Miale at University of Miami.—John D. Miale, M.D., Director of Laboratories at St. Joseph's Hosiptal, Marshfield, Wis., has been named Professor of Clinical Pathology at the University of Miami School of Medicine, Coral Gables, Fla., and Director of Clinical Pathology at Jackson Memorial Hospital, Miami, Fla.

International Association of Medical Museums.—The scientific program of the International Association of Medical Museums at its annual meeting in Philadelphia on April 6 and 7, 1954, will include a course on pathologic physiology and surgical pathology of the liver. Edward A. Gall, M.D., will serve as moderator. Discussions will cover the embryology, anatomy, physiology, biochemistry, and pathology of the liver, in addition to medical, surgical, and investigative considerations of hepatic diseases. The course will be open to all members of the association and to all pathologists, including those in training.

New Appointment for Dr. Kuzma.—Joseph F. Kuzma, M.D., has been appointed Professor and Director of the Department of Pathology at Marquette University School of Medicine. In this position he succeeds W. A. D. Anderson, M.D., who recently accepted the Chairmanship of the Department of Pathology at the University of Miami School of Medicine, Coral Gables, Fla.

Fellowships for Clinical and Experimental Research.—The establishment of two fellowships for clinical and experimental research has been announced by the Trustees of the Jewish Sanitarium and Hospital for Chronic Diseases. The fellowships, named in the honor of the President of the Hospital, Mr. Isaac Albert, will be awarded during the fall of 1953. Applicants should write to the Hospital Superintendent, Rutland Road and East 49th Street, Brooklyn.

University of Michigan.—Dr. A. James French, of the department of pathology, was recently promoted to a full professorship.

State University of New York.—Dr. Patrick James Fitzgerald has been named professor and chairman of the department of pathology of the State University of New York College of Medicine at New York City, Brooklyn. He has been assistant pathologist at Memorial Hospital and an assistant in the department of physics in the Sloan-Kettering Institute in New York. He succeeds Dr. Jean Oliver, who has held the position since 1949. Dr. Oliver has been granted special leave to engage in vital research for the Armed Forces Epidemiological Board on the renal lesion of epidemic hemorrhagic fever, a mysterious disease among the troops in Korea.

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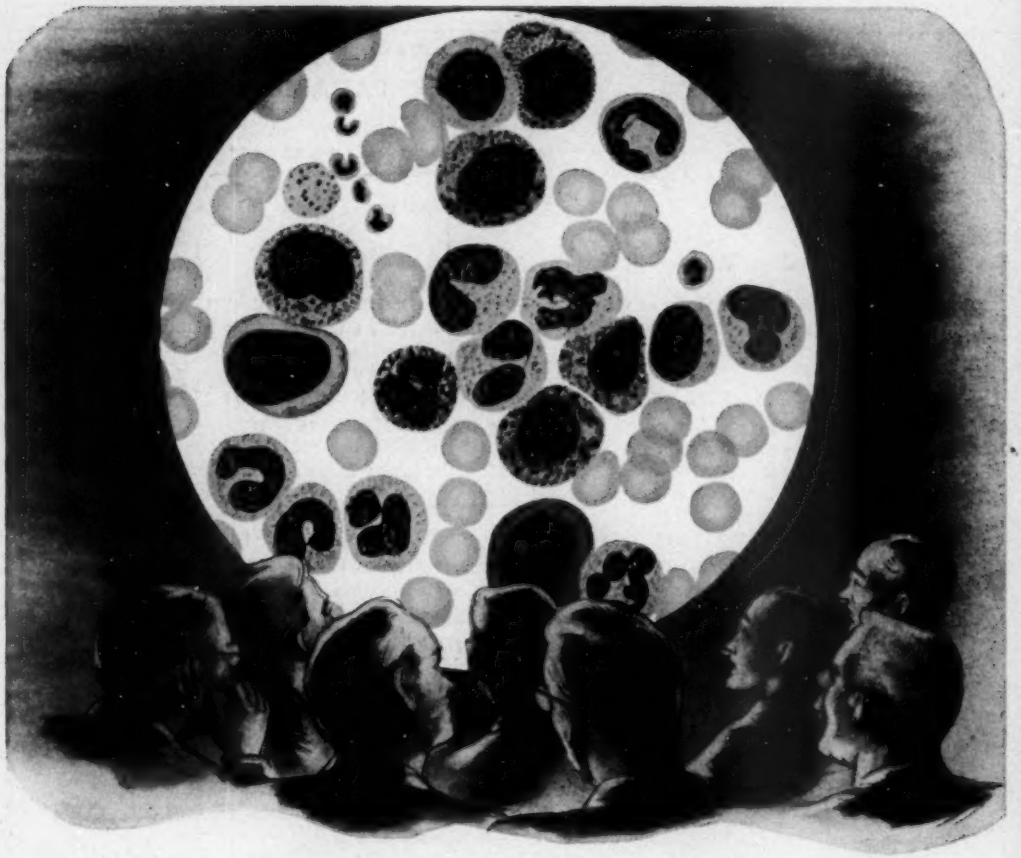

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